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### **Abstract Book of**

The 1<sup>st</sup> International Conference

## Microbial Ecology: Towards New Challenges (ICMES-2015)

Sétif, Algeria, December 12th - 14th, 2015

Organized by



### Faculté des Sciences de la Nature et de la Vie Laboratoire Microbiologie Appliquée

In collaboration of

The Tunisian Society for Microbial Ecology (ATEM)

& Laboratory of Microorganisms and Active Biomolecules, Faculty of Sciences of Tunis,

**University of Tunis El Manar** 

### **SCIENTIFIC TOPICS:**

✓ Microbial Biodiversity

- ✓ Health and Environment
- ✓ Adaptations/ Biotic Interactions
- ✓ Microbial Biotechnology and Agro-Industry



### PREAMBLE

Microbial Ecology is the study of interrelationships between microorganisms and their living and nonliving environments. It includes the study of symbioses, biogeochemical cycles and the interaction of microbes with anthropogenic effects such as pollution and climate changes. Microbial ecology is currently undergoing a revolution, with repercussions spreading throughout microbiology, ecology and ecosystem science. Understanding the ecology of microorganisms is one of the most undeniable intellectual challenges since it is essential to meet many of the major challenges facing human society today in environmental, agricultural, food, and industrial areas. Moreover, the rapid accumulation of molecular data is showing vast diversity, abundant uncultivated microbial groups and novel microbial functions which allow the study of activities of complex communities of microbes in natural, managed and engineered ecosystems.

The main objectives sought during this event is to encourage discussion and debate about the most recent advances and major issues of discipline and the sharing of knowledge and exchange of experience. It will allow researchers working in different fields of microbial ecology to meet and exchange their scientific competences and exciting results. It will also offer the opportunity for Young researchers to deepen their knowledge while expanding their horizons through the development of national and international collaborations,

This first conference on "Microbial Ecology: Towards New Challenges" has now become a need for us. The meeting is organized by the Laboratory of Applied Microbiology from the University of Sétif1, Algeria, in collaboration with the Tunisian Society for Microbial Ecology (ATEM) and the Laboratory of Microorganisms and Actives Biomolecules (LMBA) from the University of Tunis El Manar. During this conference, important and topical themes, where microbial ecology plays a key role, are addressed: i) Microbial Biodiversity ii) Health and Environment iii) Adaptations / Biotic Interactions and iv) Microbial Biotechnology and Agro-Industry.

On behalf of the Organizing Committee

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### Program of the 1st International Conference Microbial Ecology: Towards New Challenges ICMES 2015

12<sup>th</sup>-14<sup>th</sup> December, 2015

### Ferhat Abbas University, Setif 1, Algeria

Friday December 11th, 2015			
15:00-19:00 Reception and Registration at El Hidhab hotel Saturday December 12 <sup>th</sup> , 2015			
8:00-8:30			
8:00-8:30	Reception and Registration at Nait Belkacem Auditorium-UFA Sétif 1		
6.50-9.00	Opening & UFA Sétif 1 Rector's Allocution		
Session 1. B	AI: Biotechnology & Agro-industry		
9:00-9:30	Keynote 1: BAI		
	Co-Chairs: Daoud Harzallah & Abdellatif Boudebous		
	White Rot Fungi: A single tool to face multiple challenges		
	Atef Jaouani, Tunisia		
	Selected oral presentations		
9:30-9:45	Purification and structure elucidation of the active metabolites of the marine strain <i>Streptomyces sundarbansensis</i>		
0 45 40 00	Mouloud Kecha, Algeria		
9:45-10:00	Comparative study on the optimization and characterization of exopolysaccharides from <i>Kocuria rosea</i> and		
	Halogeometricum borinquense and their bioflocculant activities Habib Chouchane, Tunisia		
10.00 10.15	Statistical optimization of dilute acid hydrolysis of sawdust for lactic acid production		
10.00-10.13	Naassa Diboune, Algeria		
10:15-10:45	Coffee Break		
10:45-11:15	Keynote 2: BAI		
	Co-Chairs: Atef Jaouani & Aïcha Nancib		
	Production of Heteropolysaccharide Bioflocculant by Actinobacteria Culture Consortium and Media		
	Optimization by Surface Response Model		
	Uchechukwu U. Nwodo South Africa		
11:15-12:00	Selected oral presentations		
	Pseudomonas extremorientalis BU118: a new salt-tolerant laccase secreting bacterium with biotechnological		
	potential in textile azo dye decolorization		
	Mohamed Neifar, Tunisia		
11:30-11:45	Partial purification and characterization of tow xylanases produced by the strain Jonesia denitrificans		
	Hakim Hebal, Algeria		
11:45-12:00	The Streptomyces flora of Algeria and it's potential as a source of cellulases		
	Azzeddine Bettache, Algeria		
12:00-12:30	Poster session BAI		
12:30-14:00	Lunch		
12.30-14.00			
Session 2, HE: Health & Environment			
14:00-14:30	Keynote 3: HE		
	Co-Chairs: Belhadj Omrane & Bilal Yahiaoui		
	Unraveling the activity and diversity of Ammonia Oxidizing Microorganisms in natural and artificial ecosystems		
	Luciano Beneduce, Italy		
14.20 15.45			
	Selected oral presentations Estimation of Incidence of surgical site infections and identification of different types of pathogenic bacteria		
1	and evaluation of risk factors in orthopaedic department in Benghazi Medical Center- Libya		
	Geis Abdelsalam Ikhleif, Libya		
14:45-15:00	Isolation, Identification and Removal of Microalgae from Keddara Dam Water using Natural Bioflocculant		
	Hassiba Zemmouri, Algeria		

15:00-15:15	Characteristics of extended-spectrum β-lactamase- (ESBL) and AmpC from water samples in Tunisia beta-lactamase-producing Enterobacteriaceae isolated Leila Bensaid, Tunisia
15:15-15:30	Phenotypic and genotypic patterns of <i>Salmonella</i> isolates from human patients in two Algerian regions. Nardjess Bouzidi, Algeria
15:30-15:45	Antibiotic resistance genes and virulence factors of <i>Escherichia coli</i> isolated from birds Houssem Benyahia, Tunisia
15:45-16:15	Coffee Break
16:15-17:15	Selected oral presentations
	Co-Chairs: Abdelhakim Aouf & Rachida Yahiaoui-Zaïdi
16:15-16:30	Antimicrobial Resistance and virulence genes of <i>Staphylococcus aureus</i> from Bovine mastitis milk samples
	in Tunisia. Amira Klibi, Tunisia
16:30-16:45	Use of biochemical markers in the early detection of bacterial meningitis
	Fatima Mehenni, Algeria
16:45-17:00	First report of Extended Spectrum β-lactamases among clinical isolates of Klebsiella pneumoniae in Gaza Strip,
	Palestine
	Ghassan Tayh, Tunisia
17:00-17:15	Toxicity of ammonium based ionic liquids towards "Shewanella sp."
	Hakima Kebaili, Algeria
17:15-17:45	Poster session HE
	ember 13 <sup>th</sup> , 2015 Bl: Adaptation & Biotic Interactions
8:30-9:00	Keynote 4: ABI
	Co-Chairs: Luciano Beneduce & Imen Ouzari-Cherif
	The Frankia-Casuarinaceae nitrogen-fixing symbiosis: study of early molecular mechanisms promoting the
	symbiotic interaction and the adaptation to poor and degraded soils
	Hassen Gherbi, France
9:00-9:30	Keynote 5: ABI
	Root nodules in <i>Casuarina glauca</i> are also induced by non- <i>Frankia</i> actinobacteria of the genus <i>Nocardia</i>
	Faten Ghodbane Gtari, Tunisia
9:30-10:15	Selected oral presentations
9:30-9:45	Effects of two microbial organisms (fungus and bacterium) on the desert locust Schistocerca gregaria
	Abderrahim Khenioui, Algeria
9:45-10:00	Effects of plant growth promoting rhizobacteria associated to citrus tree to alleviate plant drought stress
40.00.40.45	Marwa Cherni, Tunisia
10:00-10:15	Image analysis and quantitative PCR method for rapid assessment of fungicide resistance in plant pathogens
	Semcheddine Cherrad, France
10:15-10:45	Coffee Break
10:45-11:15	Keynote 6: ABI
	Co-Chairs: Hassen Gherbi & Nathalie Diagne
	The impact of Arbuscular mycorrhizal fungi and Frankia on Casuarina equisetifolia growth and soil microbial
	catabolism diversity depends on the origin of the symbiotic microorganisms
	Nathalie Diagne, Senegal
11:15-12:00	Selected oral presentations
	Preview of bacterial community in the actinorhizal tree Casuarina sp. phytosphere
	Amir Ktari, Tunis, Tunisia
11:30-11:45	Physico-chemical and microbiological characterisation of steppe soils (Region of el Bayadh) in Algeria
	Fatima Zohra Abdi, Mascara, Algeria
11:45-12:00	Characterization of microorganisms associated with Algerian honeybee: Apis mellifera intermissa and Apis
	mellifera sahariensis .
	Nora Chahbar-Adidou, Algeria
12:00-12:30	Poster session ABI

Session 4, M	IB: Microbial Biodiversity
14:00-14:30	Keynote 7: MB
	Co-Chairs: Arnab Sen & Hafsa Cherif-Silini
	Culturomics, improved media prediction and phylogenomics in Microbial Ecology
	Hans Peter Klenk, UK
14:30-15:00	Keynote 8: MB
	Promises of next generation sequencing for microbial ecology
	Lassaad Belbahri, Switzerland
15:00-15:45	Selected oral presentations
15:00-15:15	Blastococcus capsiensis sp. nov., isolated from an archaeological Roman swimming pool
	Karima Hezbri, Tunisia
15:15-15:30	Identification of aerobic spore forming and non-spore-forming bacteria isolated from Tunisian milk
	Souhir Kmiha, Tunisia
15:30-15:45	Investigation of Nattrassia mangiferae Isolated from Date Palm on different Hosts
	Muntasir Adam Mohammed Elamien, Sudan
15:45-16:15	Coffee Break
16:15-16:45	Keynote 9: MB
	Co-Chairs: Larbi Larous & Lassaad Belbahri
	The story of Actinobacteria revisited in the light of Bioinformatics
	Arnab Sen, India
16:45-17:15	Selected oral presentations
	Flow cytometry approach for studying the interaction between Bacillus mojavensis and Alternaria alternata
	Asma Milet, Algeria
17:00-17:15	Preview of salt tolerant and phosphate solubilizer bacteria from date palm rhizobium and their plant growth
	promotion potentialities
	Raoudha Ferjani, Tunisia
17:00:17:15	Phenotypic and genotypic diversity of bacteria isolated from root nodules of Lucerne (Medicago sativa L.) cultivated in Algeria.
	Baha Nassima, Algeria
17:15-17:45	Poster session MB
17:45-18:00	Closure ceremony and rewarding
20:00	Social dinner
Monday. De	cember 14 <sup>th</sup> 2015

13:00 Lunch on site

09:00

Congress Tour: Djemila archaeological site

12:30-14:00 Lunch

15:00 Return to Setif

# Session 01

# Microbial Biotechnology and Agro-Industry

### Keynote 1

### White Rot Fungi: A single tool to face multiple challenges

### Atef Jaouani

Laboratoire Microorganismes et Biomolécules Actives, Faculté des Sciences de Tunis, & Institut Supérieur des Sciences Biologiques Appliquées de Tunis, Université Tunis El Manar, Tunis, Tunisia

### Abstract

White Rot Fungi are among few fungi belonging to the groups of basidiomycetes and ascomycetes which are specialized in wood degradation. They are adapted to address the major constraints of such process ie: complex composition, low nitrogen and phosphorus contents and the presence of potentially toxic compounds, thanks to their robust and versatile extracellular lignin modifying enzymes (LME). The LME system is composed of (i) peroxidases (EC1.11.1.x) that typically use hydrogen peroxide as an electron acceptor: lignin peroxidase (LiP, EC 1.11.1.14), manganese peroxidase (MnP, EC 1.11.1.13), versatile peroxidase (VP) dye peroxidase (DyP, EC 1.11.1.19) and (ii) Laccase (benzenediol (or *p*-diphenol): oxygen oxidoreductase, EC 1.10.3.2) which is a copper oxidase requiring molecular oxygen. The substrate spectrum of LME enzymes and their respective mediators comprises a wide variety of aromatic and polyaromatic compounds whatever the ring substitution (hydroxyl, alkyl, methoxyl, amine, nitrite, halogens,...etc). In addition to their unique enzymatic system, the edible nature of WRF widely open their exploitation potential. This presentation will provide a substantial contribution to the physiology, biochemistry and ecology of WRF. Recent advances of our research team to unravel the potential of white rot fungi in bioremediation, food processing and animal feed will be presented. Current studies on the biodiversity of WRF in Tunisia will also be discussed.

### Keynote 2

### Production of Heteropolysaccharide Bioflocculant by Actinobacteria Culture Consortium and Media Optimization by Surface Response Model

### Uchechukwu U. Nwodo<sup>1,2\*</sup> and Anthony I. Okoh<sup>1,2</sup>

<sup>1</sup>Applied and Environmental Microbiology Research Group (AEMREG), Department of Biochemistry and Microbiology, University of Fort Hare, Private Bag X1314, Alice 5700, South Africa. <sup>2</sup>SA-MRC Microbial Water Quality Monitoring Centre, University of Fort Hare, Alice 5700, South Africa \*Corresponding author: <u>UNwodo@ufh.ac.za</u>; tel.: +27786273279; I-fax.: +27862707453

#### Abstract

Conventionally used industrial flocculants have been implicated in deleterious and debilitating health problems hence, the need for a safe alternative. Two actinobacteria species previously identified, by 16S rDNA sequencing, as *Cellulomonas* and *Brachybacterium* species were evaluated in consortium for enhanced production of bioflocculant. Media components most suitable for the production of bioflocculants were glucose (92% flocculation activity;  $4.06 \pm 0.62$  g/l yield), peptone (89% flocculation activity;  $3.88 \pm 0.432$  g/l yield) and magnesium chloride (83% flocculation activity;  $3.99 \pm 0.79$  g/l yield) respectively. Plackett-Burman experimental design revealed glucose, peptone and magnesium chloride as critical in the production of bioflocculant with optimum concentrations 16.0, 1.5 and 1.6 (g/l) respectively achieved through central composite design. High regression coefficient ( $R^2 = 0.865$ ) shows model adequacy. FTIR spectrometry of the bioflocculant showed groups typical of heteropolysaccharide while SEM imaging revealed amorphous membranous pleated sheets with interstice of less than 1 µm. The high flocculation activity portends industrial relevance.

**Keywords:** Bioflocculant; *Brachybacterium*; *Cellulomonas*; Consortium; Heteropolysaccharide; Plackett-Burman Model.

# **ORAL COMMUNICATIONS**

### Purification and structure elucidation of the active metabolites of the marine strain Streptomyces sundarbansensis

#### Mouloud Kecha<sup>1</sup>\*, Ibtissem Djinni<sup>1</sup>, Andrea Defant<sup>2</sup> and Ines Mancini<sup>2</sup>

<sup>1</sup>Laboratoire de Microbiologie Appliquée, Faculté des Sciences de la Nature et de la Vie, Université Abderrahmane MIRA de Bejaia, Targa Ouzemmour, 06000 Bejaia, Algeria. <sup>2</sup>Laboratoire de Chimie Bioorganique Université de Trento, Italie.

\*Corresponding author: biokecha@yahoo.fr

### Abstract

Considering all the morphological, chemotaxonomic, phylogenic and physiological analyses, the name Streptomyces sundarbansensis was proposed for this marine isolate. The results obtained in this study, could be summarized as follows: Analysis of the ethyl acetate crude extracts by chromatographic techniques (liquid chromatography on PLC and HPLC), to obtain pure compounds, whose molecular structures was mainly established by mass spectrometry and NMR spectroscopy. From the cultured strain Streptomyces sundarbansensis four pure compounds were purified and structurally characterized as polyketides of the phaeochromycins family, peculiar for the presence of a propyl chain. The isolated metabolites were three known compounds (phaeochromycin B, phaeochromycin C, phaeochromycin E) and [=2-hydroxy-5-((6-hydroxy-4-oxo-4*H*-pyran-2-yl) а new molecule methyl)-2propylchroman-4-one]. In order to establish the 4-hydroxy  $\alpha$ -pyrone or 2-hydroxy  $\gamma$ -pyrone tautomeric form for these metabolites, a method was first proposed based on the assignments of absorption bands in the experimental IR spectra by the comparison with Density Functional Theory (DFT) calculated spectra for the two forms. The results indicated a  $\gamma$ -pyrone structure for the new metabolite, in line with the X-ray defined structure for the related polyketides mutactin and SEK34. Due to this study, it is possible to suggest that also the known phaeochromycins were isolated mainly in this tautomeric form, differently by the structures reported until now. Evaluation of IC50 values on the pure and structurally defined metabolites as inhibitors of Gram positive and Gram negative bacteria, from where the new compound showed the highest and selective antibacterial activity against MRSA. For the production of bioactive metabolites by strain Streptomyces sundarbansensis. It was concluded that conditions allowing a fast cell growth and biomass production are different from the ones from metabolite production conditions.

Keywords: Streptomyces sundarbansensis, antibiotics, purification, structure elucidation

## Comparative study on the optimization and characterization of exopolysaccharides from *Kocuria rosea* and *Halogeometricum borinquense* and their bioflocculant activities

<u>Habib Chouchane<sup>1\*</sup></u>, Mohamed Neifar<sup>1</sup>, Sameh Boualie<sup>1</sup>, Mouna Mahjoubi<sup>1</sup>, Afef Najjari<sup>2</sup>, Atef Jaouani<sup>2</sup>, Ahmed Slaheddine Masmoudi<sup>1</sup> and Ameur Cherif<sup>1</sup>

<sup>1</sup>Univ. Manouba, ISBST, LR11-ES31 BVBGR, Biotechpole Sidi Thabet, 2020, Ariana, Tunisia. <sup>2</sup>Laboratory of Microorganisms and Active Biomolecules, Faculty of Sciences of Tunis, University of Tunis El Manar, Tunis, Tunisia.

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#### Abstract

Biopolymers are considered a potential alternative to conventional chemical polymers because of their biodegradability, high efficiency, non-toxicity and non-secondary pollution. Recently, extracellular polymeric substances (EPS) produced by microorganisms have been recognized by many researchers as a potential flocculent for their applications in various water and wastewater treatment processes. In this context, the present work aims to optimize and characterize EPSs named pKr and pHb from two microbial isolates, Kocuria rosea (Bacteria) and Halogeometricum borinquence (Archaea), respectively. The investigation was done to determine the optimal variables of nutritional and environmental conditions to get maximum EPS based on response surface methodology. The highest bioflocculant yields of 12.3 g/L were obtained from *H. boringuence* and 3.53 g/L from *K. rosea*. A compositional analysis indicated that pKr consisted of 12% proteins and 78% carbohydrates. The composition of pHb was found to be 30.4% polysaccharides and 50.2% protein. A Fourier transform infrared spectrum confirmed the presence of carboxyl, hydroxyl, methoxyl and amino groups for both EPSs. These groups are typical for heteropolysaccharides and preferred for flocculation. Additionally, a series of experiments was performed to investigate the flocculant potential of pKr and pHb using kaolin clay at different flocculant dosage, pH of suspension, temperature, and presence of cations. Results showed that pkr and pHb were pH tolerant within rang of 2-11, with optimal activity of 89.6% at pH7 and of 83.70% at pH 11, respectively. Addition of divalent cations enhances significantly their flocculating rate more than monovalent and trivalent cations. Moreover, the flocculation performance of pKr was high and stable at temperature ranging from 30 to 100°C, within which, the minimum flocculating rate was 85.5%. However, the flocculating rate of pHb decreased by approximately 20% to 23% after heating for 15 min at 100 °C. The polysaccharide-backbone composition of pKr was assumed to explain its excellent thermal stability. On the whole comparatively, pKr and pHb demonstrated encouraging results in terms of bioflocculant activity and show potential for industrial application.

**Keywords:** Response-surface methodology, pKr, pHb, flocculating activity, pH tolerant, thermal stability.

### Statistical optimization of dilute acid hydrolysis of sawdust for lactic acid production

Naassa Diboune\*<sup>1</sup> Aicha Nancib<sup>1</sup>, Nabil Nancib<sup>1</sup> and Joseph Boudrant<sup>2</sup>

<sup>1</sup> Laboratory of Applied Microbiology, Ferhat Abbas University, 19000 Setif 1, Algeria

<sup>2</sup> Laboratory Reactions and Chemical Engineering (LRGP), UMR CNRS 7224, University of Lorraine,

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### Abstract

Lignocellulosic biomass offers a favorable alternative as a feedstock for the biological production of lactic acid because it is readily available, has no competing food value, and is less expensive than sugars as glucose or sucrose. Woody biomass is one of the potential renewable resources for the production of metabolites. The conversion of this lignocellulosic biomass to lactic acid requires hydrolysis prior to fermentation. In this study, a Lactococcus lactis subsp. lactis ATCC 19435 was used to determine the feasibility of utilizing sawdust as feedstock for the production of lactic acid. Response surface methodology (RSM) was employed for the analysis of the simultaneous effect of acid concentration, hydrolysis time and substrate loading on the glucose concentration obtained during acid hydrolysis of sawdust. A three variable, five-level central composite design (CCD) was used to develop a statistical model for the optimization of the process variables. The optimal hydrolysis conditions that resulted in the maximum glucose concentration were acid concentration; 8% (v/v), substrate loading; 16% (w/v) and pretreatment time; 2 h. Under these conditions, the maximum concentration of glucose was obtained to be 8.76 g/l. This was close to the value (8.78 g/l) obtained from experiments carried out under the optimized conditions. Lactococcus lactis subsp. lactis was found to be a potent LAB strain for the production of lactic acid utilizing steam exploded hydrolysate of sawdust and yeast extract as carbon and nitrogen sources, respectively. The final lactic acid concentration of 9.87 g/l was obtained with a volumetric productivity of 0.41g/l.h and lactic acid yield of 0.83.

**Keywords**: Wood waste, sawdust, acid hydrolysis, lignocellulosic biomass, central composite design, optimization, lactic acid.

### *Pseudomonas extremorientalis* BU118: a new salt-tolerant laccase secreting bacterium with biotechnological potential in textile azo dye decolorization

### <u>Mohamed Neifar</u><sup>1\*</sup>, Mouna Mahjoubi<sup>1</sup>, Feryel Cherni<sup>1</sup>, Habib Chouchane<sup>1</sup>, Yasmine Souissi<sup>2</sup>, Atef Jaouani<sup>3</sup> Ahmed Saleheddine<sup>1</sup> Masmoudi and Ameur Cherif<sup>1</sup>

<sup>1</sup>University of Manouba, ISBST, LBVBGR-LR11ES31, 2020, Sidi Thabet, Tunisia

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#### Abstract

Enzymatic-assisted removal of hazardous dyes is considered as a green alternative and ecofriendly method compared to those of physicochemical techniques. The present investigation focused on screening of new potent bacterial strains for laccase production and optimizing the process parameters to achieve the maximum enzymatic decolorization of textile azodye Congo red. Seven hydrocarbonoclastic bacterial strains were selected as positive in laccase production in solid medium using 2,6 dimethoxyphenol as an enzyme activity indicator. The best laccase producer Pseudomonas extremorientalis BU118 showed a maximum laccase activity of about 7000U/L under optimal conditions. The influence of different concentration of dye, enzyme, salt and incubation time on Congo red decolorization was studied using central composite design and response surface methodology to find the optimum conditions required for maximum decolorization by P. extremorientalis laccase. The enzyme exhibited a remarkable color removal capability over a wide range of dye and salt concentrations. The phytotoxicity of laccase-treated dye was significantly reduced compared to the untreated dye. The above results introduce P. extremorientalis laccase as an efficient halostable biocatalyst for decolorization of Congo red, a diazodye. Further decolorization studies should be performed on dyes from other families such as triphenylmethan, indigo, and anthraquinone dyes, extensively employed in textile industries.

**Keywords:** *Pseudomonas extremorientalis* laccase, azo dye decolorization; salt tolerance, response surface methodology; Central composite design

### BAI-05

### Partial purification and characterization of tow xylanases produced by the the strain Jonesia denitrificans

#### Hakim Hebal \*., F. Hamzaoui and S. Benallaoua

Laboratoire de Microbiologie Appliquée, Faculté des Sciences de la Nature et de la Vie. Université A/Mira de Béjaia, Targa Ouzemmour 06000, Algérie.

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### Abstract

Ligniocellulose is the most abundant and renewable material in the world. Its use in biotechnology to produce value added products, require enzymes like cellulases and xylanases. However, these technologies use harsh conditions (high temperature, high pH pressure and salinity), and these enzymes must be active and stable in these conditions to sustain the production. Xylanases are important enzymes in this production as xylane is the most abundant polymer after cellulose. In this study, tow xylanases were partially purified from Jonesia denitrificans and their main proprieties characterized. The production of xylanases was optimal at 37°C and pH 9,5. The enzymes were purified by ammonium sulfate precipitation and gel filtration chromatography. The SDS-PAGE gel shows tow xylanases of 35 kDa (XYNA) and 120 kDa (XYNB). The enzymes are optimally active at 50°C and pH 7 for XYN A, and 50° and pH 6 for XYNB. The xylanases are extremely halotolerant, with an optimal at 5% NaCl, and 50% residual activity at 30% NaCl. Their tolerance to NaCl depends on the pH of activity. XYNA is more tolerant at pH 5 and XYNB at pH 8. The enzymes are also tested in presence of ionic liquids, substances that dissolve ligniocellulosic material. Their tolerance also depends on the pH of activity. With these properties, the two xylanases are susceptible of use in biotechnology.

Keywords: Xylanases, halotolerance, ionic liquids, Jonesia denitrificans.

### The Streptomyces flora of Algeria and its potential as a source of cellulases

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### Abstract

Cellulose is the most abundant organic compound in the world and it is a kind of renewable energy that human beings can easily utilize. Microbial degradation of celluloses involves the synergistic action of three major enzymes: endo-  $\beta$  -1,4-glucanase, exo-  $\beta$  -1,4- cellobiosidase and  $\beta$ -glucosidase. *Streptomyces* species are able to degrade many macromolecules such as proteins, cellulose, starch, lipids, and chitin. For the degradation of cellulsoe, hemicellulose, and lignin that are abundant in plants, different strains of *Streptomyces* sp. have been studied and found to be good producers of cellulases. Thus, the present investigation describes the production, purification and characterization of endoglucanase from *Streptomyces* sp. B-PNG23. The highest endoglucanase activity was obtained in a medium comprised of wheat bran (10 g/l), yeast extract (2 g/l), NaCl (2 g/l), NH4Cl (2.5 g/l), and (0.4 g/l) of MgSO4. *Streptomyces* sp.B-PNG23 produces five endoglucanases. The Endo2 was purified and characterized after three consecutive purification steps using ultrafiltration, anion exchange chromatography and sieving molecular. The molecular weight of the Endo2 was estimated by SDS-PAGE revealing that the isoenzyme is a monomeric enzyme of 66 kDa.

Keywords: Production, Purification, Endoglucanase, Streptomyces, Characterization.

# POSTERS

### Antibacterial activity of lactic acid bacteria strains isolated from camel and cow milk

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### Abstract

Due to antibiotic resistance phenomena menacing the public health, the studies are nowadays focused on researching natural antimicrobial substances. Lactic acid bacteria are known by their antimicrobial effects, that's why they are used in food industry as conservators. In this study, the analysis of raw camel milk from l'Oued and M'sila and of fermented cow milk collected from Sétif had led to the isolation of 76 strains of lactic acid bacteria identified on the basis of their 16S rDNA sequence as belonging to the following genera: Enterococcus, Lactobacillus, Lactococcus and Leuconostoc. Antibacterial activity experiments by the "Spot on the lawn" method, showed 26 strains only exhibiting an anti-Listeria potential from which 12, isolated from camel milk, had an important inhibitor activity (26.5 mm against Klebsiella pneumoniae). The potential high anti-Listeria activity of these lactic acid strains and their large spectrum of antibacterial activity, including spoilage bacteria (13 strains), make them good candidates in industrial food bio preservation and biotechnological transformation of milk.

**Keywords:** milk, lactic acid bacteria, antibacterial activity, bio conservation.

# *Lactobacillus plantarum* and olive oil as bio-preservation agents of a short repined goat cheese

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### Abstract

In recent years, there is a constant need for cheese having enhanced organoleptic and sanitary qualities. The aim of our study is to find a new way to ensure the preservation of short ripened goat milk cheese using a strain of Lactobacillus plantarum and virgin olive oil. The study of antibacterial activity of L. plantarum using the method of spots revealed inhibitory activity against five pathogenic strains, the most representative of the species found in milk and dairy products. Goat cheeses were developed using two strains of *Lactococcus lactis* and including the strain of L. plantarum as repining and bio-preservation strain. Manufactured cheeses were then repined at 12°C / 7days. At the end of the ripening period, diced cheese were kept in sterile glass jars containing virgin olive oil for a further 7 days at room temperature, protected from light. Monitoring of cheeses made including L. plantarum after storage in olive oil revealed no mold growth, or adverse changes in macroscopic appearance unlike manufactured cheeses without L. plantarum and which have not been retained in the olive oil. Microbiological analysis of cheese has revealed a gradual decrease of the total flora and staphylococci during preservation in olive oil. However, no effect was detected on the development of lactic acid flora including L. plantarum. These results lead to deduce that the use of the strain of L. plantarum having an antagonistic power against unwanted germs and maceration in olive oil can provide new approaches ensuring the preservation of goat cheese.

Keyword: goat cheese, bio-preservation, L. plantarum, olive oil.

### Study of Listeria monocytogenes in some food products

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### Abstract

*Listeria monocytogenes* is a Gram-positive type of bacteria responsible for listeriosis. This serious infectious disease affects both humans and animals. It is very resistant that can multiply in atmospheres having a temperature between  $1 \circ C$  and  $45 \circ C$ . The interest of our research is to enable detection of *Listeria monocytogenes* in foodstuffs affected or not by national regulations. During the study, a total of 174 samples of several types of food were examined. *Listeria monocytogenes* in food was conducted according to ISO 11290-1 and 11290-2. 45 *Listeria* strains were isolated on selective media (chromogenic agar agar or PALCAM) from several food samples mainly of red meat and derivatives (16 strains), fish and fish products (3 strains), cheeses and dairy products (11 strains) and ready meals (15 strains). The isolates were characterized morphologically and cultural microscopic observation and observation of the colonies aspect, biochemically by conventional identification tests and the gallery Api Listeria. It appears from our results the presence of four of the six species of the Listeria genus, with a predominance of *L. monocytogenes*.

Keywords: Listeria monocytogenes, human health, food, microscopic observation, Api gallery.

### Contribution to a discriminating evaluation of the microbiological quality of Tizi-Ouzou's beef meat

#### Hamida Akli \*, Boussad Metna, Sarah Lammi, Yacine Lamrous & Djamal Djenane

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### Abstract

The objective of this study was to investigate, in actual production conditions, the variability of the microbiological quality of beef meat in Tizi Ouzou's region (Algeria). A total of 373 butcheries was sampled using a stratified random sampling method where the stratifying factor constituted the five meat hygiene inspections in this region, namely the inspection of Azazga, Larbaâ Neth Irathen , Draâ El Mizan, draâ ben khedda and that of Tizi Ouzou. The sensory quality of 42 samples of the *Longissimus lumborum* muscle, was performed on two sensory characteristics of beef meat. The odor and discoloration of meat were evaluated over an interval of every other day during 34 days. In addition, the physico-chemical and microbiological analyses were carried out to complement the sensory approach. The results show a significant variability (p <0.05) of the microbiological quality through the five meat hygiene inspections. This leads to the conclusion that the factors influencing this quality are heterogeneous across the region of Tizi Ouzou, in particular the activity of the meat hygiene inspections that would be the most likely factor which explain better the resulting variability.

Keywords: Beef Longissimus lumborum, microbiological quality, stratification, Tizi Ouzou.

### The Study of the survival of some probiotic lactic acid bacteria in simulated gastrointestinal conditions after encapsulation

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### Abstract

The most important probiotic microorganisms which deeply conducted with the human gastrointestinal tract are Lactic acid bacteria (LAB). While several methods have been used to enhance the viability of probiotic bacteria, encapsulation is the best used method to immobilize, protect, and control cells. The objective of this work is to investigate the effect of microencapsulation on the viability of some probiotic lactic acid bacteria of different origins in simulated gastrointestinal tract: Lactobacillus plantarum isolated from Qlila: Algerian traditional cheese, Lactobacillus rhamnosus and Lactobacillus satsumensis isolated from goat. In our study, encapsulated cells coated by 1.6% of sodium alginate grew well in both gastric (enzymatic barrier and acid 2h/37°C) and intestinal conditions (bile salts, pancreatine 2h/37°C). The results of viable counts determined by a pour plate method using MRS agar and incubated at 37°C for 48h were expressed in percentage of viability. Results showed that encapsulated cells gave more viability than free cells (75% with Lb.rhamnosus, Lb. satsumensis with a percentage of 100% in gastric conditions). For free Lb plantarum cells we have a clear decrease in the gastrointestinal conditions at the time of 2h and 4 h. However, the encapsulated cells showed a preservation of their viability in the gastric conditions and a slight decrease in the intestinal juice by 1% after 4 h.

Keywords: probiotic bacteria, simulated gastrointestinal tract, microencapsulation.

### Antimicrobial potential of *Melia azedarach*

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#### Abstract

Melia azedarach L. (Meliaceae), deciduous tree native to northern India, was naturalized in tropical and subtropical countries. This tree was discovered for the first time to the early twentieth century, during the great locust Palestine in 1915. India in 1926, Algeria in 1888-1894. In the present work, we studied the antimicrobial potential of aqueous phenol and fats extracted from seeds and almonds of Melia azedarach L cultivated in Algeria. Phenolic compounds were extracted with distilled water as described by Oomah et al, (2008) and assayed according to the method described by Oomah, et al. (2010, 2011). Total fat of seeds and kernels were obtained by Soxhlet extraction for 6 hours with hexane as a solvent and have been the subject of a determination of yield. The antimicrobial effect of two strains of Pectobacterium was evaluated by the agar diffusion method. Various phenolic extracts of Melia azedarach induce inhibitions of growth of the two strains of Pectobacterium tested, with seed extract which exhibits the highest inhibitory capacity and which expresses the highest levels of total phenols, flavonoids and tannins (11,39 mg CE / g DM, 0.66 mg EQ / g DM and 6.15 mg CE / g DM, respectively). The results of antibacterial activity showed generally efficacity against the growth of the tested Pectobacterium, with different sensitivities wich shows a greater sensitivity of Pectobacterium carotovorum subsp atrosepticum (Pca) to fat seeds (14.41 mm) and almond (12.12 mm) compared to Pectobacterium carotovorum subsp carotovorum (Pcc).

Keywords: Melia azedarach, phenolic extracts, fatty acids, Pectobacterium

### Isolation of extremely halophilic bacteria able to produce biosurfactant

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### Abstract

Biosurfactants are surface active agents with broad range commercial applications in various industries and have considerable advantages over their chemical counterparts. In this study, drop collapse assay, oil spreading assay, emulsification assay and surface tension measurement were compared for their reliability and ease of use to screen biosurfactant producing six extremely halophilic bacteria isolated from saline soil of Chott El Hodna-M'sila (Algeria), which is considered as a thalassohaline environment. Results from screening methods revealed that, CH2 and CH5 strains are potential candidates for biosurfactant production. Among these, there was a very strong negative linear correlation between the emulsification index and surface tension (rs = -0.987) and a strong negative linear correlation between the drop collapse method and surface tension (rs = -0.807), suggesting that strains highly active in any one of these methods were active in other three methods. As every method has its advantages and disadvantages, a combination of different methods is appropriate for a successful screening.

**Keywords:** Biosurfactant, halophilic bacteria, drop collapse assay, oil spreading assay, emulsification assay, surface tension.

# Selecting producing fungi lignocellulolytic enzymes in the soil and their interest in the agro-industry

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Abstract

Lignocellulosic biomasses are the most abundant renewable resources on Earth, A large fraction of lignocelluloses is represented by residual biomass such as agro-industrial wastes, that would represent the key response to the need of increasing renewable energy production. The majority being considered wastes, its hydrolysis is performed by cellulase, hemicellulase and ligninases. This work is part of the operating movement of lignocellulosic wastes to produce enzymes cellulolytiques.63 mould strains were isolated in the region of Akbou Bejaia Algeria. Screening of cellulase activity is performed on all strains; isolates capable to grow on agar medium in the presence of CMC are subjected to a first semi-quantitative screening by the method Lugol.A second quantitative screening on liquid medium was used to select the most efficient isolates 93,65% of the isolates show activity endoglucanase between 0.107-4.202 U/ml and 90.47% of these isolates have xylanolytic activity between 0,113-5,033 U/ml. 04 isolates were selected for their potential to produce interesting endoglucanases and xylanase compared to other strains or between 2,786 -4,202 U/ml and 4.763-5.033 U/ml, respectively. They are tested for their fermentative capacity to produce lignocellulases from Agro-industrial wastes such as wheat straw, wheat bran and olive grignon.

Keywords: Mould, Screening, Endoglucanase, Xylanase.

## Impact of the acid stress on the strains of *Lactobacillus* isolated from south algerian camel milk

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### Abstract

The practices of dairy industries cause variations of physicochemical properties of the microorganisms used, including lactobacilli. These microorganisms have developed responses to address these variations. Our work consisted in studying the acid stress response in *Lactobacillus* strains isolated from southern Algerian camel milk. We exposed cells of *Lactobacillus* to acid stress by growing them at different pH (pH 1, pH 2, pH 3, pH 4, pH 5, pH 6). Growth and the number of viable and culturable cells were determined at various times. The behavior of cells adapted to a moderate acid shock was evaluated in the same way, and by determining viable culturable cells. The results showed that the growth parameters, the viability of the cells and the mortality rates are significantly affected as the distance of the optimum pH for growth. The adaptation of the cells has a positive effect on the parameters examined except at extreme pH (pH 1 and pH 2). The exposition of the cells to an acid stress affects cell proliferation and survival of *Lactobacillus*. The adaptation of the cells to an acid stress affects were parameters by making them more resistant to the stress.

Keywords: Lactobacillus – Stress – Acid – Adaptation – Survival.

## Solid state fermentation of *Bjerkandera adusta* for manganese peroxidase production on agro residues

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### Abstract:

Manganese peroxidase is the most common lignin- modifying peroxidase produced by almost all wood-colonizing basidiomycetes causing white-rot and various soil-colonizing litterdecomposing fungi. This study presents the screening of Bjerkandera adusta BRFM 1916, a white-rot fungus, isolated from an Algerian forest, for its ability to produce extracellular oxidases enzymes on solid medium containing 2, 2'-azino-bis-[3-ethylthiazoline-6-sulfonate] (ABTS) and guaiacol. The strain showed ABTS - and guaiacol-oxidation activities indicating the presence of ligninolytic enzymes. Several substrates for manganese peroxidase (MnP) production by Bjerkandera adusta BRFM 1916 were screened using solid state fermentation. The olive cake followed by wheat bran, as a carbon source, were found to be the most efficient substrates compared to wheat straw, miscanthus, grape pulp, barley bran, miscanthus/olive cake (50/50) (M/OC) and wheat straw/olive cake (50/50)(WS/OC). The maximum MnP activity was found to be around 1200 U/L and 1049 U/L with olive cake and wheat bran respectively, while 872, 762, 341, 323, 258 and 107 U/L were obtained from M/OC, WS/OC, wheat straw, barley bran, miscanthus and grape pulp respectively. Moreover, the activity in M/OC substrate was 3.37 times higher than the activity obtained with the miscanthus whereas WS/OC gives 2.23 times higher than WS. This result shows that the olive cake improves MnP secretion mixed with straw or miscanthus. Of eight lignocellulosic residues tested, olive cake and wheat bran provided the highest MnP activity.

Keywords: *Bjerkandera adusta*, Manganese peroxidase, solid-state fermentation, agro residues.

### Probiotic potential of Lactobacillus strains isolated from fresh bee pollen

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### Abstract

Lactic acid bacteria have received much attention over recent decades due to health promoting properties of certain strains, called probiotics and the increasing consumer that diet and health are linked. The aim of the present study was to evaluate potential probiotic of twenty strains of Lactobacillus isolated from fresh bee pollen grains. Therefore, Lactobacillus strains were studied for their antibacterial activity against four pathogenic bacteria Salmonella typhimurium ATCC 13311, Escherichia coli ATCC 25922, Staphylococcus aureus ATCC 25923 and *Listeria innocua* CLIP 74915, their ability to survive under conditions simulating human gastrointestinal tract was investigated at low pH and in the presence of bile salts. Moreover, their safety was verified by testing haemolytic activity on human blood agar and antibiotic resistance. The results showed that all lactobacillus strains were effective against all indicator bacteria; the most sensitive was Staphylococcus aureus, the highest diameter of inhibition zone was 48mm observed against Salmonella typhimurium. 50% of strains could survive with losses in cell viability after 3h exposure to pH=2, while all strains were able to maintained their viability after 3h to pH=3 and 4h in the presence 0.3% of bile salts. All lactobacillus strains were sensitive to Chloramphenicol, Nitroxoline, Cefotaxime, Penicillin G, Amoxycillin, Pristinomycin and Cefexim. No one showed a haemolysis on blood agar. Our Results showed that the Lactobacillus strains exhibited a remarkable inhibitory action against both Gram positive and negative pathogenic, the *Lactobacillus* LB11 and LB12 were found to possess good probiotic properties, they present an important interest for industrial use.

Keywords: Lactobacillus, Probiotic, Survival, bee pollen, antibacterial activity.

### Olive waste fermentation and hydrolytic enzymes production by rhizospheric isolate of *Pseudomonas fluorescens* D1

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### Abstract

In the last two decades, hydrolytic enzymes are gaining increased attention for their potential in biotechnology processes, such as biomass bioconversion into fermentable sugars, for ethanol production which increase the demands to find more efficient hydrolytic enzymes producing microorganisms from different biotopes aquatic, terrestrial or rhizospheric. In this context our present study was conducted to isolate, screen and evaluates the enzymatic activities of rhizospheric microflora of the following plants: the argan (Argania spinosa L), olive (Olea europaea L), retame (Retama monosperma L), the spider plant (Chlorophytum comosum L) and mauve (Malva sylvestris L). The results of the selective isolation and enzymatic screening on cellulose, pectin and starch based media, reveal the existence of 22 isolate( 48%) amylolytic, 14 (30%) cellulolytic and 10 (22%) pectinolytic). , as well as the purification on potato dextrose agar (PDA) for fungi and nutritive agar (NA) for bacteria followed by Macroscopic and microscopic examinations indicate that the 46 microbial isolates were (72% bacteria and 28% fungi) with a moderate microbial diversity associated with argan and olive rhizospheres noticed for amylolytic, pectinolytic and cellulolytic respectively. Rhizospheric isolates of Pseudomonas fluorescens were subjected to successive rounds of selective purification on King B medium and enzymatic screening, the strain Pseudomonas fluorescens D1 was chosen as potent candidate on its hyper and multiple enzyme activities, the potent strain was tested for olive wastes degradation and hydrolytic enzymes production via grounded olive kernels lequid state fermentation for two weeks incubation at 30°C. The results show significant biomass degradation, olive oil recovery, and multiple enzymes production (cellulase, pectinase, amylase, lipase and peroxidase) assessed through well cut diffusion technique. The promising results obtained by the rhizospheric Pseudomonas fluorescens D1 and grounded olive kernels make them the good combination for olive wastes recycling and multiple enzyme production for useful application in the agrofood sector.

**Keywords:** olive wastes; rhizosphere; microorganisms; *Pseudomonas*, fermentation; hydrolytic enzymes.

## Study of antimicrobial effect of PVDF/PMMA blend on growth inhibiting of microorganisms

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### Abstract

Growth of microorganisms on different materials may cause aesthetic and structural damage. Due to the structure of PVDF, it exhibits an excellent chemical resistance, an easy processing and an excellent UV stability; but disadvantages such as inability to produce a glossy finish and the high cost, make it indispensable to introduce a secondary polymer. The PMMA has a low price compared to PVDF but it deteriorates easily by UV radiations exposure; this character limits its use as protecting agent for stones. Blending the two polymers gives not only good compatibility with P but also provides good heat resistance, mechanical properties, weather ability and optical clarity [1]. After blending these two polymers in the DMF, films are obtained to evaluate the blend resistance against bacterial flora. It was found that the infrared spectroscopy (FTIR) of PVDF/PMMA films showed the superposition of the spectra of all compositions, with the exclusion of any chemical reaction between the two polymers or the presence of the double bonds characteristic of PVDF deshydrofluoration. The UV visible spectroscopy, before and after exposure to artificial weathering, showed that the PVDF's absorbance values are invariants (about 200 nm after two years of the exposition to the aging test). In contrast, the absorbance of PMMA has changed at the same wavelength, which explains its tendency to degradation.

Keywords: Inhibiting effect, PVDF/PMMA blend, Microorganisms, FTIR, U.V, Spectroscopy.

## The highlighting of laccase activity produced by *Trametes sp.* and *Chaetomium sp.* On medium pumpkin waste based

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### Abstract:

The objective of this work is to demonstrate the laccasic activity produced on a medium containing pumpkin waste by two fungal strains belonging to the genus *Trametes sp.* and *Chaetomium sp.* The development test of *Trametes sp.* et *Cheatomium sp.* revealed that these strains having the ability to grow and produce laccase by using pumpkin waste as medium. Indeed, the fermentation of liquid medium without adding any inducer showed that laccase activity obtained is more important than that obtained after the addition of  $CaCl_2$  as an inducer. So the production of laccase without the inductor is more important than that with the inducer (CaCl<sub>2</sub>).

Keywords: laccase activity, pumpkin scrap, Trametes sp., Chaetomium sp., liquid medium.

### Effect of domestic and industrial pasteurization on raw goat milk

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### Abstract

Milk whatever type is an important product for all consumers because of composition. It suitable to direct research works to compare commercial pasteurized goat's milk present in our markets and commercial raw goat's milk taken by farms and pasteurized at home. For that purpose, comparative steps were realized between these milks allowing of this fact their highlighting. This comparison leaned essentially on the bacteriological analysis. Physical-chemistry and nutritional analyses were including too with parameters: density measure, pH, acidity, protein and lipid quantities. The result of this study revealed an acceptable hygienic quality, except coliforms. The technological parameters have a satisfactory aspect. Concerning the biochemical composition, it was turned difference because food's nature of goats.

Keywords: goat's milk, pasteurization, hygienic quality, nutritional quality.

### Flocculant properties and dye removal potentiality of a new glycoprotein from the Archaeal strain Halogeometricum borinquense

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### Abstract

The bioflocculant production, characterization and potential application in dye removal by the newly isolated *Halogeometricum boringuense* from Tunisian desert were investigated. The application of the response surface methodology allowed us to determine a set of optimal conditions (glucose, 20 g/L, casaminoacids, 15 g/L, Nacl concentration, 20% and incubation time, 8 days) for bioflocculant production. Experiments carried out under these conditions led to bioflocculant yield of 12.52 g/L. Chemical analyses of the bioflocculant showed 30.4% carbohydrate and 50.2% protein. Fourier transform infrared spectroscopy (FTIR) indicated the presence of carboxyl, hydroxyl amino and amide groups, amongst others, typical for glycoprotein. To further study the flocculating activity, factors such as bioflocculant dosage, temperature, pH of the reaction solution and cations addition were tested. The highest flocculating rate of kaolin clay was 83.70% obtained at bioflocculant concentration of 0.20 mg/l, pH of 11 and temperature of 50 °C. The current study found that addition of divalent cation such as  $Ca^{2+}$  and  $Mn^{2+}$  enhances the flocculating rate and the glycoprotein is a negatively charged bioflocculant. The effectiveness of such bioflocculant in the decolorization of 8 dyes was then examined. The glycoprotein flocculant was effective in flocculating some soluble anionic dyes in aqueous solution, in particular Reactive Blue 4 and Acid Yellow with a decolorization efficiency of 78.8% and 74.6%, respectively. The decolorization efficiency was dependent on the flocculant dosage and solution pH. H. boringuense appears to hold promise as a source of new bioflocculant that could stand as an alternative to inorganic and synthetic organic flocculants.

**Keywords:** glycoprotein bioflocculant – response surface methodology -flocculation activity - dve decolorization - Halogeometricum boringuense.

## Partial characterization of xylanases from *Bacillus oceanisediminis* SJ3 isolated from Algerian soil

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### Abstract

A new Bacillus oceanisediminis SJ3 strain which produces thermostable xylanases was isolated from Algerian soil, it has been selected as a promising strain for xylanase production. This latter was 20.24 U/ml after 2 days in the presence of oat spelts xylan. The xylanase activity was characterized in terms of temperature and pH profiles and thermostabiliy and effect of the metal ions and solvents. The results indicated that the enzyme was optimally active at pH 7.0. The optimum temperature for xylanase activity was 55°C. At this temperature, the half life was of 22 h and the enzyme retained 50% of its activity after its incubation for 2.67 h at 100°C. These properties qualify the enzyme to be highly thermostable and potentially important for application in some industrial processes such as bioconversion of lignocellulosic materials into fermentative products. The detergent SDS and the  $\beta$ mercaptoethanol were strong inhibitors, while all the ions tested do not affect the enzyme activity. Xylanases are, at least, stable in all organic solvents tested except for propanol where a reduction of 46.5% was observed. Zymogram analysis and SDS-PAGE of the culture supernatant indicated the presence of four bands suggesting the presence of four isoenzymes (xyl a, xyl b, xyl c and xyl d) with respective molecular weight of : 162 ; 117,5 ; 83,7 and 59 KDa.

Keywords: Bacillus oceanisediminis SJ3, thermostable, xylanase, oat spelts xylan.

### Theoretical study of the interactions involved in the inhibition of *Mycobacterium tuberculosis* methionine aminopeptidase by several molecules

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### Abstract

The main purpose of this study was to test the performance of certain molecular docking programs that simulate the interactions between proteins and ligands. To study these interactions, we chose two molecular docking programs: Surflex, which uses incremental method and GOLD, which uses a genetic algorithm. In this context, we have used these programs to study the interactions involved in the inhibition of methionine aminopeptidase by various inhibitors. The latter is a potential therapeutic target in *Mycobacterium tuberculosis* infection. In the first part of our work we tested the two programs Surflex and GOLD according to the RMSD between the predicted mode of interaction and the crystal structure. Both programs, Surflex and GOLD can be considered as sufficiently effective reproduce quite well the experimental results with over 66% of the values of RMSD below 2 Å for the former and 79.16% for the second. In the second part of this work we have tried to contribute firstly, understanding the basic mechanisms of the bond between a target protein and its ligand and the other in search of potential therapeutic agents by molecular docking. For this, we studied the inhibition of MetAP by various molecules from the literature and also visualized the links they involve the active site of this enzyme. The results are generally similar for both programs Surflex and GOLD. The study showed that the compound TO7 (Affinity =  $3.10 \text{ M}^{-1}$ , Fitness = 57.35) has the best inhibitory effect of MetAP enzyme.

Keywords: Molecular Docking, Surflex, GOLD, RMSD, Methionine Aminopeptidase, *Mycobacterium tuberculosis*.

# Assessment of technological traits of autochthonous thermophilic starters isolated from Algerian traditional dairy products

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### Abstract

Algeria is the first consumer of milk and dairy products in Maghreb (3 billion liters a year). Nevertheless, it remains dependent in matter of supply with lactic starters against the foreign laboratories and makes resort to the importation of these products for dairy products manufacture. The aim of this study is to evaluate various technological traits of three autochthonous thermophilic starters isolated from ovine traditional butter and curd milk, compared to an industrial one. The acidification aptitude was estimated by measurement of pH and titrable acidity. Moreover, exopolysaccharides production was tested in hypersaccharose agar, the proteolysis was evaluated in nutritive broth supplemented with sterile reconstituted skim milk, and A/B agar. The products of proteolysis were determined using HPLC. Lipolysis was estimated using stab culture on; triglycerides agar, and MEVAG medium with 1% of triglyceride. The different starters were individually grown in reconstituted skim milk and whole milk (28% fat) to determine their abilities to produce acetaldehyde. With the exception of the acidification activity which was considered to be weak (2.3g/L, 2.35g/L 4.15g/L/4h with 3% of culture), the obtained results indicated that starters have a good texturing aptitude (66g/L, 124g/L, 136g/L these of exopolysaccharides/24 h) and proteolytic ability (0.1 tryptic U) which led to liberate Lproline and glycine in the medium by two starters, they have also a capacity to degrade lipids contrary to commercial starter. The remarkable presence of acetaldehyde in the cultured milk (8.2 mg/L, 9.6 mg/L and 10.2 mg/L) confirmed their aptitude to synthesize aromatic substances.

**Keywords:** Autochthonous thermophilic starters, technological traits, acidification, proteolysis, acetaldehyde.

# Research of the proteolytic potential in strains of *Bacillus* genus isolated from protein rich soils

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# Abstract

In this work 22 different strains belonging to the genus *Bacillus* characterized by their performance degrade proteins were isolated from 15 soil samples rich in protein. The study of the proteolytic activity produced on milk agar at a concentration of 10% skim milk, showed that 5 strains gave very low hydrolysis zones (<5 mm), whereas 10 strains gave hydrolysis areas having diameters varying between 10 and 20 mm. Of these, six strains (diameter> 10 mm) were selected. The production of protease by Bacillus strains 6 select was carried out on two different media, TGEA or NG medium containing 10g/l skim milk. *Bacillus thuringiensis* and *licheniformis* are the most powerful strains. 5 strains are selected for biochemical identification, and *B. thuringiensis* the most efficient strain was selected for optimization of culture conditions. The maximum production of protease was obtained after incubation for 48 h in the presence of 1% glucose, 1% peptone. A significant reduction of the production was observed in the presence of 1.5% peptone.

Keywords: protease, *Bacillus*, proteolytic activity, agar milk.

### Screening of lipid accumulating microorganisms on agro industrial by product

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### Abstract

Rapid fluctuations in prices of basic resources and the climate change associated with increasing greenhouse gas emissions have stimulated the development of alternative sources for biomass based energy fuels such as biodiesel. However, this bioenergy renewable source is mainly derived from vegetable oils produced on agricultural land which limits the largescale development of this technology. To face with this situation, several alternatives have been identified namely the use of microorganisms such as bacteria, micro-algae, fungi and yeasts for the production of lipids. The nitrogen limitation is the most efficient condition for inducing lipogenesis and consequently accumulation of cytosolic lipid droplets. First, the isolation of efficient lipid accumulating bacteria and yeasts were carried out from various habitats i.e. olive oil mill wastewaters and oil factory residues. Fifty (50) oleaginous strains were successfully isolated based on "Sudan Black" and "Nile Red" screening methods. Isolates were identified by sequencing 16S and ITS regions for bacteria and veasts, respectively. So far identified bacteria belong to the genera Terribacillus, Bacillus, Pseudomonas, Enterobacter, Pseudocitrobacter, Shewanella, Lysinibacillus, Pantoea whereas yeasts belong to the genera Rhodotorula, Candida and Meyerozyma. Optimization of the production of lipids of the selected isolates is currently undertaken on carbon rich nitrogen deficient agro industrial by products.

Keywords: Oleaginous microorganism, lipid accumulation, Nile Red, Sudan black B, Biodiesel.

### Effects of tanniferous plants on rumen microbial profile of cattle

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### Abstract

Plant rich secondary compounds have antimicrobial effects by acting against different rumen microbial populations. The objective of this study was to determine the effects of fodder shrubs (*Acacia nilotica* and *A. cyanophylla*), rich in secondary metabolites on the metabolic activity of rumen microbiota of cattle. The qualitative and quantitative determination of in *vitro* fermentation gases was used as a marker of fermentation substrates. The monitoring of quantitative fluctuations in the equilibrium of the major populations of rumen microbiota was performed by the real-time PCR technique. Molecular analysis revealed important changes in the microbial profile of the rumen: a significant imbalance among cellulolytic groups, including, *Fibrobacter succinogenes* population which increased relatively with all the substrates, unlike *Ruminoccocus flavefaciens* species which decreased significantly, suggesting a selective effect of tannins on the latter. A significant anti protozoan effect of shrub species was also observed in parallel to the inhibition of the production of ammonia. Furthermore, the tannins content of *Acacia* had antimethanogenic activities but without, apparently, direct effects on relative abundance of methanogenic *Archaea* populations, indicating a potential resistance of this population to tannins of the studied species.

Key words: Acacia, in vitro fermentescibility, rumen microbiota, real time PCR, tannins.

# Caracterisation and evaluation of the effect of cold storage and industrial aromas on the viability of Bifidobacteria in fermented dairy products

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### Abstract:

Bifidobacteria are the most important group of probiotic cultures usually incorporated in fermented dairy products. Lots of technological aspects have to be considered when selecting probiotic strains. This study aims to assess the effect of two very important of those factors which are the cold storage at 4°C and the contact with synthesized aromas used in dairy industry. Bifidobacteria strains were isolated on MRS medium (added of 0.05% cysteine-HCL and 2 mg/l of nalidixic acid) from different origins (breast-fed infant feces and two probiotic yoghurts produced and commercialized in Algeria). The viability during cold storage has been evaluated in skimmed milk for 21 days, accompanied by a measurement of post acidification for the same period. While the effect of industrial aromas has been followed using MRScys broth added of 0,2% v/v of two aromas used in dairy industry, (banana and vanilla). The isolated strains belong to the following species: Bifidobacterium longum, B. breve and B. animalis. The studied strains retain good viability after 7 days of storage at 4 °C up to 58%, but a significant loss of viability was noted along the cold storage period arriving to a rate of 27.7% after 21 days. This was accompanied by a verry weak post acidification produced by the totality of studied strains which is a verry important technological character. The same strains showed a different behavior when exposed to the two aromas, with a viability from 14,4% to 81% at the end of the storage period.

**Key words:** Bifidobacteria, dairy products, technological aspects, breast-fed infant, probiotic yoghurts.

### Pollution and waste treatment through the ability of stem related to *Rhodococcus* genus

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### Abstract

Sixty strains were isolated from various terrestrial and aquatic sites. Among these, after a primary screening, based on morphological, physic-chemical, biochemical and biological characteristics, 34 strains were related to the genus *Rhodococcus*. 25 strains showed a catabolic activity towards crude oil, highlighted on solid MSM media, vaporized by the pollutant (petroleum). Another study is performed on liquid MSM and nine strains showed a degradation capacity (training biosurfactant). The level of biodegradation was estimated by the INT test. The calculation of the emulsion, shown in index strains, has outstanding values of E24. Furthermore, 19 strains showed a catabolic activity on cholesterol. This activity is likely due to cholesterol oxidase, an enzyme controlled by the CHO gene. The PCR amplification resulted in an amplification product for two laboratory control strains (*Pseudomonas*) and for a *Rhodococcus* genus related strain (LBTP2). We optimized the DNA extraction from *Rhodococcus* genus related strains and performed a PCR to amplify the 16S rDNA gene of 1500 bp.

Keywords: Rhodoccocus, biosurfactants, INT, E24, cholesterol oxidase, PCR, 16S rDNA.

# Sanitary quality and Characterization of lactic acid bacteria isolated from rural traditional Cheese (*Jben*) of Djelfa province

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# Abstract

Five samples of traditional cheese (Jben) collected in rural area of Djelfa, Algeria were studied by determination of morphological, cultural, physiological and biochemical characteristics. Among 104 isolates 29 lactic acid bacterial (LAB) strains were isolated and purified. This study shows that the biodiversity of this traditional cheese studied is characterized by enterococci, lactobacilli and lactococci. The most common lactic acid bacteria belonging to the species Lactococcus lactis (10.34%), Lactococcus raffinolactis (6.90%), Lactococcus cremoris (06.90%), Lactobacillus Plantarium (13.79%), Lactobacillus rhamnosus (10.34%), Lactobacillus fermentum (10.34%), Lactobacillus acidophilus (13.79%) ), Lactobacillus casei (10.34%), Lactobacillus helveticus (06.90%) and other species of the genus *Enterococcus* (10.34%). The samples pH average was 4, 42  $\pm$  0.15, mean titratable acidity value was  $79.4 \pm 3$ , 11° D and microbiological analysis results were ; total mesophilic aerobic bacteria 6.12 x 10<sup>6</sup> UFC/g, coliform bacteria 1.04 x 10<sup>4</sup> UFC/g, Yeast 1.08x10<sup>4</sup> UFC/g. Staphylococcus, Salmonella and moulds weren't detected. In our work, we tested the acidifying power, proteolytic activity and the antimicrobial effect of these LAB isolated against Staphylococcus aureus. The action of the bacteriocins was eliminated by proteolytic enzymes.

Keyword: Jben, Lactic acid bacteria, bacteriocins, acidifying power, proteolytic activity,.

# Production of siderophore and indol acetic acid by actinomycetes from extreme

### environments (sebkha soil)

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### Abstract

The aim of this study was to isolate actinomycetes from salt soil (sebkha) and investigate their capacities to produce siderophore and the plant growth hormone indole-3-acetic acid (IAA). Soil samples were collected from different sites of sebkha soil of Bazer (Sétif, Algeria) in the North East of Algeria to isolate actinomycetes strains in the first on starch casein agar (SCA). Population enumeration, phenotypic characters like substrate mycelium, aerial mycelium, aerial mass colour, reverse side pigment, biochemical characteristics were also studied and finally siderophore production by inoculating the grown isolates in CAS agar and IAA production in SCA medium supplemented with 2 mg/mL of L-tryptophan. From 34 isolates, 4 strains showed a positive result for siderophore production by the formation of orang halo around the colony and 5 actinomycetes strains isolated were effective in producing IAA by different concentrations from10.54 $\mu$ g/ml to 17.93 $\mu$ g/ml. This study indicated that some actinomycetes strains from extreme environment had the potential to produce siderophore and IAA, which can be better exploited to be used as prospective bio control agents in agriculture system.

Keywords: Actinomycetes, salt soil, siderophore, IAA, PGPR, bioactive.

# Improving the Catalytic Properties of Immobilized Lecitase from *Thermomyces lanuginosus/Fusarium oxysporum* via physical adsorption onto bentonite

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### Abstract

The current study reports on an immobilized phospholipase A1 onto bentonite. The enzyme, a microbial lipase was produced by submerged fermentation of *Thermomyces lanuginosus* and *F. oxysporum*. This enzyme was immobilized onto unmodified bentonite which was applied for soybean oil degumming. The immobilized enzyme exhibits a novel behavior: It displays high phospholipase and lipase activities at neutral pH, while the maximum phospholipase activity was found to shift to 55 °C and both enzyme activities were enhanced by a factor of 2 to 10 than its maximum normal activity. In terms of specific activity, immobilized PLA<sub>1</sub> had a specific activity equaled to 80% of its free form. Moreover adsorption of PLA<sub>1</sub> onto the bentonite was found to be effective in reducing phosphorus contents from soybean oil.

Keywords: Submerged fermentation; Lecitase; Immobilization; Physical adsorption.

# Application of a novel keratinase Purified from submerged culture of *Actinomadura keratinilytica cpt20* in the biodegradation of feather wastes

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# Abstract

The present presentation reports on the purification, biochemical characterization and application of a novel extracellular thermoactive keratinase (KERB) newly isolated from *Actinomadura keratinilytica* strain Cpt20. The sequence of the N-terminal residues of KERB showed high homology with those of Actinomycetes keratinases. After purification, Optimal activity was achieved at pH 8 and 50°C, and KERB was highly stable at broad pH and temperature range (6–10) and (30–50  $\circ$ C), thermoactivity and thermostability were upgraded in the presence of 10 mM Ca2+. The enzyme was completely inhibited by phenylmethanesulfonyl fluoride (PMSF) and diiodopropyl fluorophosphates (DFP), which suggests that it belongs to the serine protease family. The enzyme also exhibited powerful keratinolytic activity that made it able to accomplish the entire feather-biodegradation process on its own, which provides strong support for the potential candidacy of this enzyme as an effective and eco-friendly alternative to the conventional chemicals used for the biodegradation of keratin-containing wastes.

# Study of the proteolytic activity of microorganisms with high probiotic potential, involved in industrial biotechnology

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### Abstract

Lactic acid bacteria are used empirically for centuries in the manufacture and bioconservation of many fermented foods such as vegetables, meats, and as starters in the manufacture of yoghurt. The interest of this work is to target local strains of lactic native flora of arid and semi-arid areas of Algeria with interesting technology skills. The identification of bacteria necessarily involves observation of phenotypic and physico-chemical characteristics that lead to define gender in most cases These analyzes must be supported by genetic testing to provide a more-limited taxonomic position. This work allowed to process 55 samples of raw camel milk, goat, sheep and traditional butter. He focused mainly on the isolation of potentially probiotic thermophilic strains. In total 37 strains were isolated including 20 strains of thermophilic streptococci and 17 strains of *Lactobacillus delbrueckii* group. These strains have very valuable technology skills such as the study of the proteolytic activity, lipolytic,  $\beta$ -galactosidase, acidifying power, aromatics production, antibacterial activity vis-a-vis pathogenic strains, including *Enterococcus faecalis JH2 -2, Escherichia coli*. Furthermore DNA pairs of primers used for PCR-based amplification has given a reliable identification of *Streptococcus thermophilus*.

Pro A do 5'-CCC GTC TCA TCC AAA CTG TG-3' Pro B up 5'-TGG TTC AAG TTA GGG CCT CA -3'

It is necessary to remember that local lactic acid bacteria can eliminate most of the pathogenic bacteria, as it would be interesting to offer these probiotic bacteria as starter culture for the production of fermented foods et.la bioconservation.

**Keywords:** DNA, proteolytic activity, antibacterial activity,  $\beta$ -galactosidase activity, aromatic compounds.

# Extraction and quantification of pectate lyases enzymes from potato tissues infected by pectinolytic bacteria belonging to the genus *Dickeya* sp.

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### Abstract

Potatoes are the main target of a large group of pathogenic organisms. Among them, species belonging to the genera Pectobacterium/Dickeva induces important economic losses. These bacteria are characterized by great maceration activity due to the production of pectinolytic enzymes. The aim of our study is to highlight the presence of enzymes during infection. For this, a sample of 11 cultivars was used. Tubers were infected by 3 strains of Dickeya "2 strains of D. dadantii : wild type (470), mutant type (350) and a strain of D. solani) ". A standard inoculum was injected to half-tubers, then 100µl are taken from symptomatic tissues and put into selective medium. Cultures were incubated at 30° C for 48 hours under gentle agitation. After incubation, 0.5ml was mixed with 5ml medium. And after centrifugation, supernatant is recovered and enzymes are quantified by spectrophotometry at 230 nm. According to the results, we find that mutant strains 350 produces the most quantities of Pels, followed by D. solani 99, while wild strain 470 is the least productive. It was also found that among varieties used, pectate lyase activity occurs especially on cultivar Monalisa, Charlotte and Franceline, which is in accordance with the results of infection test. Whereas, Pels are under expressed in Agata and Ditta varieties, who recorded the lowest rate of enzymes. This study demonstrates the potential infectivity of *Dickeya*, as well as the response of potatoes varieties to the enzyme synthesis.

Keywords: Dickeya sp, potatoes, pectate lyases, enzyme assay, pathogenicity.

### Microbial removal of o-cresol from water using a pure strain of Pseudomonas sp.

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### Abstract

Due to the toxicity of *o*-cresol, efficiently removing of this compound by bacteria is of great importance. *o*-cresol is a characteristic pollutant in wastewaters and effluents from oil refineries, pesticides, dyes, resins and petrochemicals production. It is listed as a priority pollutant by the agency for Toxic Substances and Disease Registry (ATSDR, 2007) and U.S. Environmental Protection Agency (EPA). Biodegradation is a promising alternative, effective and simple of biological treatment. It represents an environmental friendly procedure and there is an increasing interest in isolating and identifying microorganisms with phenolic compounds metabolizing capacity. Indeed, despite the toxicity of cresols, some microorganisms such as bacteria can tolerate and use these compounds as energy/carbon source, even at relatively high concentration. The present study deals with the removal of *o*-cresol in water using biodegradation by pure strain of *Pseudomonas* sp. isolated from soil in Algeria. The kinetic of *o*-cresol biodegradation by this strain was investigated in batch reactors for an initial *o*-cresol concentration ranging from 250-1250 mg.L<sup>-1</sup>.

Keywords: Pseudomonas sp., o-cresol, biodegradation, kinetic, reactors.

# Isolation and identification of some *Bacillus thuringiensis* strains with insecticidal activity against *Ceratitis capitata*

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# Abstract

The aims of the present work is to study the effect of toxins (delta-endotoxins), extracted from different strains of *Bacillus thuringiensis* on *Ceratitis capitata*, a devastating of citrus and fruit trees. Strains of *B. thuringiensis* were isolated from the mud of Sebket Sejoumi. Among 70 isolates tested, 15 showed a significant identicalness activity in which 5 isolates led to mortality rates  $\geq 90 \%$ . These mortality rates are caused by endotoxins of *B. thuringiensis*. Analysis of proteins profiles of different isolates of *B. thuringiensis* revealed variability between them. The preliminary results of this study encourage us towards the characterization of the insecticidal activity produced by *B. thuringiensis* strains for large scale application.

Keywords: Bacillus thuringiensis, insecticidal activity, endotoxins, Ceratitis capitata.

# Screening and production of polyhydroxyalcanoates by *Bacillus megaterium* using beet molasses as carbon sources

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### Abstract

The extensive usage of petrochemical plastics due to their versatile properties especially durability is causing severe problem in waste management affecting the aesthetic quality of cities, water bodies and natural areas. Polyhydroxalcanoates (PHAs) have been recognized as a desirable alternative material to the non-biodegradable petrochemical produced polymers. In the present study, an attempt was made to screening a potential polyhydroxyalcanoates accumulating Bacillus strains isolated from different grass soil samples in Western Algeria and optimization of media parameters for increased PHA production by strain able to use agro-industrial waste products (Sugarcane and beet molasses) as carbon source. Fifty-two Bacillus isolated strains were screened for intracellular lipids by staining with Sudan Black B. Of these, 8 isolates were positively stained. The strain L9 based on morphological and physiological properties, and sequence of its 16S rDNA it was identified as Bacillus megaterium, this strain was explored for a potential to synthesize PHA using different Carbon and nitrogen sources. The accumulation of biopolymer granule in cells of Bacillus megaterium strain L9 significantly depended on the ratio of carbon and nitrogen sources in culture. The highest PHA production was observed with 3% of beet molasses after 48h of growth (41% w/w), whereas the highest biomass (0,6 g.L<sup>-1</sup>) was obtained at 4% beet molasses supplemented with 0,05% ammonium chloride. The present study gives insight into benefits of conversion of available agro-industrial wastes materials into a valuable product.

Keywords: Polyhydroxyalcanoates, Bacillus megaterium, beet molasses.

# Effects of selected medicinal plants on ruminal fermentation, fibre digestion and methane production *in vitro*

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### Abstract

The objective of this in vitro study was to compare fermentation patterns of three selected traditional medicinal plants and to evaluate their potential as antimethanogenic additives in ruminant feeds. Effects of Nigella sativa, Rosmarinus Officinalis and Zingiber Officinale, which were incorporated to the fermentation substrate as a dry powder, on ruminal fermentation, fibre digestion and methane production were studied in vitro in batch cultures of mixed rumen microorganisms. Serum bottles containing 500 mg of substrate (500 g alfalfa hav/kg, 400 g grass hav/kg and 100 g barley grain), 50 mg of the plant additive tested and 50 ml of buffered rumen fluid (10 ml sheep rumen fluid + 40 ml culture medium) were incubated at 39 °C for 24 h. After incubation, gas and methane production, pH and volatile fatty acid (VFA) concentration in the incubation medium and dry matter and neutral detergent fibre disappearance were recorded. In general, a high methane reduction was noted with Monensin (control) and Nigella sativa and no effect was observed with the other treatments. The amounts of methane produced after 24 hours of fermentation varies between 0,28 and 1,12 mmol/g DM. Monensin drives the highest percentage reduction, with a value of 75% and Nigella sativa recorded a 20% reduction in value. This study should be complemented by other *in vitro* investigations to determine the dose effect of Nigella sativa and also studies by the Rusitec to ensure stability in time of the observed effects.

Keywords: in vitro fermentation, methanogenesis, medicinal plants, ruminants.

# The influence of environmental factors and *Debaryomyces Hansenii* on growth of *Fusarium moniliforme*

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### Abstract

*Fusarium* is a phytopathogenic fungus with a global distribution, capable of infecting a wide range of crop plants, were it cause several diseases and produce mycotoxins responsible for serious chronic and acute disease for human and animals. In this study the influence of temperature (5-40°C), pH (4-9), water activity (aw), and culture media (PDA, MEA, YESA, Czapek), and carbon source (glucose, sucrose, starch) were tested on growth of *F. moniliforme*. The in vitro assays showed that the growth was maximum at 25°C and pH=5. The exposure of the fungus to the 90% relative humidity resulted maximum mecylial growth of *F. moniliforme* compared to the other levels. The radial growth was more abundant on PDA than on MEA, YESA, and Czapek. The growth was optimal in presence of glucose as carbon source. In biological control test, the antagonistic agent *Debaryomyces hansenii* was effective in reducing the linear growth of pathogen by their secondary metabolites, volatiles and non-volatiles compounds, mycoparasitism, and competition for space and nutriments.

Keywords: *Fusarium moniliforme*; Environmental factor; Culture media; Water activity; Temperature; biocontrol; *Debaryomyces hansenii*.

# A halotolerant endospore forming Bacilli, producing antimicrobial compounds and industrially important enzymes

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### Abstract

This study concern the analysis of an endospore forming bacterium LMB9832 able to grow at NaCl concentration range of 0-25 %, isolated from an Algerian hypersaline environment "El Golea lake" for antimicrobial compounds and industrial enzymes biosynthesis. Antimicrobial activities were extracted by different solvent to choice the best one for extraction. The most active crude extract was analyzed by thin layer chromatography on silica and Infra-Rouge spectra. The results showed that LMB9832 possess an important antimicrobial potential, it is active against Gram positive and negative bacteria and Fungi. The chloroform is the best solvent to extract her antimicrobial activities. TLC and Infra-Red spectra make clear the presence of two types of molecules: amines and phenols. LMB3982 was able to produce variety of enzymes and utilize a number of subtracts (gelatin, starch, casein, tween80, lecithin and esculin). All results reveal a real biotechnological trump of the strain. Such strains are researched in biotechnology because of their antimicrobial power, their hydrolytic activities and their power to bear the stress physicochemical conditions. Molecular characterization is necessary to identify the strain and characterization of bioactive compound by developed techniques such as HPLC, column chromatography and mass spectroscopy, is also necessary to know their chemical nature.

Keywords: Halo-tolerance, Bacilli, Antimicrobial compounds, Enzymes

# Protection of buffalo skin products from bacterial degradation using metalloproteinase inhibitors

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### Abstract

Deterioration of fresh hides and skins due to microbial growth and activity is a major problem in the leather and tanning industry and a major cause of economic loss. Since finished leather is primarily collagen, damage to skins and hides caused by proteolytic bacteria during stages of processing animal skins and hides is particularly harmful and adversely affects the quality of the finished leather. While biocides or preservatives may work well in inhibiting or killing microorganisms, they are generally not effective to block collagenase enzymes which can remain active even after biocide treatment. Present investigation aims to provide a method for protecting buffalo skins and hides from collagenases produced by microorganisms that come into contact with the skins and hides during storage and/or leather tanning process in Tunisian leather industry (TMM). Collagenase producing bacteria were firstly screened from different untreated and treated buffalo skin samples using nutrient agar amended with azocoll. Enzyme activity was revealed by visual inspection of the clear zone around the bacteria in the azocoll medium. Thirty isolates were then chosen for cultivation in liquid media supplemented with 1% collagen as a sole carbon source and the amounts of enzyme produced were analyzed. The collagen degrading isolates selected on the basis of their high collagenase activities were identified phylogenetically by 16S rRNA analysis. Inhibition of bacterial collagenases was performed by a metalloproteinase inhibitor (disodium ethylene diamine tetra acetic acid, EDTA) at different inhibitory concentrations. This efficient inhibitor may be applied directly by spraying the solution onto the animal skin or hide.

Keywords: Buffalo hide; leather; bacterial degradation; proteolytic enzymes; collagenase inhibitor

# Effect of *Rhizobium* and PSB inoculation on *Pisum sativum* growth and symbiotic relation in two types of soils

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### Abstract

A pot experiment were conducted at the Faculty of Agriculture, Omdurman Islamic University, Sudan to study the effect of Rhizobium leguminosarum by. viceae (TAL1399 -EN9) and phosphorus solubilizing bacteria (PSB) Bacillus megaterium var. phosphaticum (Phosphobacterin – Phosphorin) inoculation and their interaction on the growth of pea (Pisum sativum) in 2 types of soil (soil A and B). The experiment was carried out by using complete randomized design. The pea parameters were nodulation and shoot, root dry weight. Soil A silty loam (pH = 8.1, EC = 1ms/cm, SAR = 0.65meq/l, N = 0.084%, P = 0.008%) and soil B sandy loam (pH = 7.52, EC = 48ms/cm, SAR = 1.25meq/l, N = 0.101%, P = 0.01%). In soil A, phosphobacterin significantly (P≤0.05) increased nodulation, shoot and root dry weight compared to control. The highest results on nodule dry weight were observed by the coinoculation of TAL1399 with phosphorin and phosphobacterin respectively. EN 9 + phosphobacterin gave the highest shoot dry weight at 8 weeks after sowing (WAS) compared to control. In soil B, plants germination and growth was very weak and there were no nodule formation at the whole time of experiment. After 8 WAS all plants were lost. Co-inoculation of TAL1399 + phosphorin significantly (P≤0.05) increased root dry weight at 4 WAS and inoculation with phosphorin alone at 6 WAS compared to control and other treatments. Moreover, inoculation with EN9, co-inoculation of TAL1399 + phosphobacterin respectively gave a significant increment on the average of shoot dry weight compared to control.

Keywords: bacteria, co-inoculation, nodulation, pea.

# Enzymatic activity of endophytic actinomycetes and fungi isolated isolated from Agricultural crops

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# Abstract

The objectives of this work are to isolate endophytic actinomycetes and fungi from wheat (*Triticum durum* Desf), and screening of some extracellular enzymes of biotechnological interest such as in the valuation of agricultural and agro-industrial residues, processing of foods, manufacturing of detergents, textiles, pharmaceutical products, medical therapy and in molecular biology. Three actinomycetes and seven fungal isolates were isolated from the leaves and roots and identified at genus level based on the morphology of the fungal culturein a previous study. The productions of following enzymes were analysed: amylase, esterase, lipase, cellulose and protease by radial diffusion in solid media method.As results it was observed that all the isolates possessed at least one enzymatic activity tested especially the three isolates of actinomycetes, which was ranked as follows: (100%) of them have amylase activity, proteolytic (90%), lipolytic (50%), and esterasic (70%). while, cellulolytic activity was detected (40%) of isolates. This study is a preliminary qualitative test for extracellular enzymes production from endophytes and from these results, it is revealed that endophytes are able to represented a new source with applicable biotechnological potential in different areas such as in the nutrition, detergent, paper, pharmaceutical, textile and leather industries.

Key words: endophytes, enzyme production, Triticum durum Desf., actinomycetes, fungi.

# Non polyenic antifungal molecules produced by a salt tolerant and alkaliphilic actinobacteria, *Streptomyces* sp. BS30, isolated from an arid soil of Algeria

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### Abstract

Under study and exploration of new unusual ecosystems to isolate new strains of actinobacteria probably producing new bioactive molecules, we looked for arid soils of the Algerian Sahara. The actinomycete strain designated BS30 was selected for its remarkable antifungal and antibacterial activity; antifungal activity against Aspergillus genus and phytopathogenic species. Analysis of the 16S rRNA gene sequence (1 479 nucleotides) assigned this strain to Streptomyces genus with 98.77% similarity with Streptomyces *spiroverticillatus* NBRC 12821<sup>T</sup>, the most closely related. Extraction of antifungal molecules was performed from liquid cultures. Two bioactive molecules against the pathogen fungi Aspergillus Niger are produced by strain BS30, the first extracted with dichloromethane and the second with the *n*-butanol. The fraction extracted with dichloromethane has molecular weight 245 g / mol and a maximum absorption under UV at 365 nm. The second fraction obtained after extraction with *n*-butanol has molecular weight 411.32 g / mol. It is slightly colored in yellow, soluble and stable in methanol, and has a maximum absorption under UV at 200 nm. The chemical characterization allowed the classification of these two molecules in the aromatic antibiotics family. The two molecules are not polyenic antifungal; they are original and different from existing antifungal molecules, which suggest that they are new antifungal molecules.

Keywords: Arid soil; Actinobacteria; Antifungal; non polyenic; 16S rRNA

# Crude oil biodegradability distillate and naphtha B by consortium and its pure strains isolated from the wastewater treatment plant of Skikda refinery

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### Abstract

Elimination of petroleum products of the marine environment requires the involvement of various biotic and abiotic factors. Among these factors, the biodegradation by microorganisms. The aim of our work is to isolate and identify microbial strains of the effluents of the treatment plant (ETPII), in refinery Skikda (RA1K), then test their petroleum biodegradation capacity and its distillate in a synthetic sea water, and their ability to produce biosurfactants. A consortium and other five microbial strains were selected and identified : *Bacillus sp., Micrococcus sp., Arthrobacter sp., Alcaligenes sp.,* and a fungi *Geotrichum candidum.* The obtained results showed that the microbial consortium is more active than the individual strains with a biodegradability percentage of 45% for the Naphtha B and 37.62% for the crude petroleum after 3 weeks of incubation. Comparing the percentage of biodegradability of the two carbon sources, mentioned that: *Bacillus sp.> Micrococcus sp.> Ceotrichum candidum> Arthrobacter sp. > Alcaligenes sp.* The results of the emulsion indexe show that there is a correlation between the biodegradability percentage and emulsion percentage of the crude petroleum. This confirms that the biosurfactants are factors favoring petroleum biodegradation.

Keywords: Biodegradability, the Consortium, Bacillus, emulsion Index, crude petroleum.

# Screening of Antimicrobial Activities of the Endophytic Fungi isolated from *Rhamnus alternus*

#### Nouari Sadrati<sup>1</sup>, <u>Amina Zerroug<sup>1</sup></u>\*, Daoud Harzallah<sup>1</sup>, Sabrina Bakli<sup>1</sup> & Younes Saadoune<sup>1</sup>

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### Abstract

From a pharmaceutical point of view, there is a growing need for new antibiotics, chemotherapeutic agents, and agrochemicals that are highly effective, possess low toxicity, and have a minor environmental impact. Endophytes are microorganisms that reside asymptomatically in the tissues of higher plants and are a promising source of novel organic natural metabolites exhibiting a variety of biological activities. The present study was conducted to evaluate the antimicrobial activity of endophytic fungi isolated from roots, branches and leaves of the medicinal plant Rhamnus Alternus, where colonization percentages obtained were 57.5 %, 35%, 7.5 % respectively. The antimicrobial of 40 endophytic fungi isolated from R. Alternus were screened against six pathogenic bacteria, three are Gramnegative, three are Gram-positive and against three phytopathogenic fungi by dual culture method. All isolates showed inhibitory activity on at least one or more pathogenic bacteria, wherein the zones of inhibition ranged from 0 to 30 mm, the latter was obtained by the isolate Alternaria sp.5 against Bacillus cereus. For the inhibition percentages of phytopathogenic fungi were ranged from 0 to 80.76 %, the latter was obtained by the isolate Scytalidium sp. against Phytophthora infestans. These results have allowed us to select the active isolates and identified to genus level, and the dominant genera were Alternaria sp., Ulocladium sp., Curvularia sp., Phoma sp., Aureobasidium sp., Scytalidium sp., Onychocola sp., Paecilomyces sp., Talaromyces sp., Acremonium sp., and some strains with sterile mycelium.

Keywords: Endophytic fungi, medicinal plants, *Rhamnus Alternus*, colonization rates, antimicrobial activity.

### Application of indigenous and exotic arbuscular mycorrhizal fungi on industrial tomato

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### Abstract

Mycorrhizal fungi (AMF) are the most prevalent plant-microbe symbiosis. The interest of using such fungi for sustainable agriculture is increasing due to their role in improving the health of plants, soil fertility and stability of aggregates. Tomatoes like most crop plants can form symbioses with AMF. An experiment in controlled conditions was conducted to evaluate the effect of selected native AMF species and exotic species of commercial inoculums on the growth of a variety of industrial tomato (Lycopersicon esculentum Mill. var. Isma F1) grown in Northeast Algeria. The plants were inoculated with a fungal isolate composed of a mixture of spores of two native species retrieved from Algerian soils (Funneliformis mosseae and Glomus constrictum) and with a commercial mycorrhizal inoculum, composed of 6 exotic species. A soil from a farm field previously cultivated of wheat and planned for the cultivation of tomato, containing all the microflora and the same sterilized soil were used as growing media. After two months of growth, the results showed that mycorrhizal inoculation improved significantly some growth parameters such as height, fresh weight of shoot and root parts, and the dry weight of the shoot parts. Generally, commercial inoculum and local fungal isolate had a similar effect on growth. Our results affirm that there is a positive interaction between introduced and indigenous AMF and that extracted AMF from Algerian soils can replace commercial mycorrhizal inoculum, and may offer economically and ecologically important advantages in sustainable or organic cropping systems.

Keywords: Arbuscular mycorrhiza, exotic inoculum, native AMF, growth, tomato.

# Fungal diversity in Saharan Oases in Southern Tunisia

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### Abstract

Extreme environments are often exposed to harsh conditions such as high temperature, pressure and the fluctuations of nutrient sources that make these sites ideal candidates for the study of microbial diversity. Oases are isolated areas of vegetation in desert where the date palm forms a key plant providing ecological services. In addition to their benefits, date palm (*Phoenix dactylifera L.*) influences the diversity of mycorhizal fungi and shapes their community structure due to a selective pressure alongside abiotic factors. The present study was undertaken to assess the diversity of the fungal community in different fractions soil particularly in the Tunisian desert palm oases by culture-dependent approaches. Samples from (i) soil surrounding the date palm roots and (ii) uncultivated soil (bulk soil) were collected from three different Oases: Ksar ghuilane, Douz and Nafta. Morphological and microscopic identification of fungal isolates were confirmed by sequencing of the ITS region and the amplification of specific regions such as beta.-tubline, Actin and Calmoduline. This polyphasic approach has resulted in a final identification for 102 strains that distributed into 52 species and 27 genera with a dominance of Ascomycota (*Aspergillus, Penicillium, Talaromyces, Fusarium*).

# Cultivation of macromycetes mycelium and exploration of their biotechnological potential

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### Abstract

Mushrooms' picking and cultivation would ensure to inhabitants of mountainous rural areas a substantial income. Indeed, macrofungi are widespread in Tunisian forests. However, their diversity and ecology remain largely unexplored which hindered their exploitation and valorisation. In particular, the cultivation of the mycelial form will be useful in order to: (i) facilitate the conservation fungi (ii) help the carpophore production (iii) study more in depth their diversity by molecular tools, and (iv) assess their biotechnological potential as reservoir of active biomolecules (enzymes, antioxidants,..etc). To attain these objectives, more than 50 expeditions to Tunisian forests were organized. Several hundreds of specimens were collected and macroscopically and microscopically identified. Mycelial cultivation of newly collected specimens allowed obtaining 20 isolates of basidiomycetes. Molecular analysis by sequencing ITS regions showed that the isolates belong to the following genera: *Pleurotus* (4), *Lentinus* (3), *Ganoderma* (3), *Gymnopilus* (3), *Panus* (2), *Polyporus* (2) *Coriolopsis* (2) and *Macrolepiota*. On the other hand the cultivated species were screened for their enzyme production on solid and liquid media: laccase, cellulase, pectinase and xylanase. Potentially interesting produced enzymes will be purified and characterized.

### Diversity and distribution of Macrofungi in Tunisian forests

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### Abstract

Macrofungi are important biological components of forest ecosystems . They are considered as valuable resources of (i) ecological (ii) socio-economic, (iii) biotechnological, (iv) medicinal and pharmaceutical interest. Indeed, they play an important role in the functioning of forest ecosystem namely in lignocelluloses degradation and mycorrhizal symbiosis. Moreover, they would ensure to foresters a substantial income by picking or producing edible mushrooms. On the other hand, they constitute a huge reservoir of bioactive molecules (hallucinogens, enzymes, antibiotics, anti-tumor, anticancer) of biotechnological and medicinal interests.In Tunisia, about 12% of the country area is covered by forests potentially harboring high macrofungal biodiversity. However, very little is known about their distribution, abundance and diversity. The main objective of the present work is (i) to assess the diversity of macromycetes in Tunisian forests and (ii) to set up the first Tunisian collection of macromycetes. Over 50 sampling expeditions were organized to sampling from the governorates of Jendouba, Beja, Bizerte, Nabeul, Zaghouan and Tunis. More than 700 specimens were collected. Ongoing polyphasic identification based on their macroscopic, microscopic and molecular techniques showed high fungal diversity in the studied areas. In particular, results showed higher macrofungal richness in the northwest regions of Tunisia, especially Jendouba and Beja. One hundred and twenty-three species (117 basidiomycetes and 6 ascomycetes) belonging to 78 genera and 46 families were recorded. An online database of collected specimens is under construction.

# Screening and study of antimicrobial activity of Actinomycetes isolated from extreme environments

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# Abstract

The production of new bioactive molecules is currently a major concern worldwide due to the proliferation of pathogenic microorganisms have developed resistance to existing molecules. In this context, 23 Actinomycetes strains isolated from 2 different sites (soil of Sebkha Melghir and rocks of Djemila) are the subject of this study. The macroscopic characteristics showed diversity, according to the morphology of the colonies and theirs pigmentations (gray, white, light brown or pink). The most of them have a granular appearence. Thus, the microscopic examination revealed that all the strains were Gram-positive, filamentous and spore-forming. The 16S r DNA sequence revealed that the majority belong to *Streptomyces*, Microbaterium and Kocuria The antibacterial activity was carried out against ten bacteria (Staphylococcus aureus (MRSA), Enterococcus feacalis, Staphylococcus aureus ATCC 25923, Listeria ivanovii, Pseudomonas aeruginosa ATCC 27853, Escherichia coli ATCC 25922, Proteus sp., Serratia marcescens, Salmonella sp., Enterobacter cloacae), 13 strains (56.52%) of them showed antibacterial activity against these bacteria or more using the GSM medium and the agar cylinder method. However, the antifungal activity is tested for 4 fungi (Fusarium solani (FS), Fusarium oxysporum fsp. albedinis (FOA), Phytophthora infestans (PI), Botrytis cinerae) and a yeast (Candida albicans), 20 strains (86.96%) of them have antifungal activity in the limit. these results confirm that the Streptomyces always remain a prolific source of antibiotics molecules.

Key words: bioactive molecules, Streptomyces, antibacterial activity, antifungal activity.

### Biocontrol potential of *Bacillus* isolated against phytopathogens

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### Abstract

Development of biopesticides for the biocontrol of plant diseases is a promising solution that helps fight against phytopathogenics while reducing the use of chemicals. Microbial biopesticides are much more environmentally compatible than chemicals and have an increased specificity to pathogens against which they are directed. Among the bacterial strains used as biopesticides Bacillus predominate. By their intrinsic properties with beneficial effects for plants and their ability to form spores, Bacillus strains are best suited for application to field. In this context, this work consists to evaluate the biocontrol activities of 35 strains of *Bacillus* isolated from different arid regions of Algeria. The ability of strains was evaluated by the determination of antifungal activities (volatiles and diffusibles metabolites) against phytopathogenic fungi (Botrytis cinerea, Fusarium solani and Phytophthora infestans...) and the production of extracellular enzymes (cellulase, chitinase and protease). All strains had at least one antagonistic activity against a fungal strain with a percentage of inhibition ranging from 87 to 100%. Extraction of the active principle on TSB medium of two selected strains was achieved by a Rotavapor. The extract from the D5 was antibacterial. However, the extract of D13 strain was rather antifungal. Analysis of metabolites done by thin layer chromatography showed a single compound of peptidic nature in the D5 extract. The D13 extract could contain several peptide compounds. The percentage of strains having the ability to produce extracellular enzymes efficient against fungi was important: chitinase (92.5%), protease (90%) and cellulase (42%). Several strains (n = 14) had the ability to secrete three enzymes. The most efficient strains were D4, D6, D7, D12 and represented a future path for production of antimicrobial metabolites.

Key words : Bacillus, antagonism, biocontrol, extracellular enzymes.

# Evaluation of enzymatic antagonisme capacity of some *Trichoderma* strains against *Fusarium* sp. as a phytopathogen isolated from an agricultural Algerian soil.

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# Abstract

The present study aimed to fight against the phytopathogen fungus *Fusarium sp.* Eight fungi from the genus Fusarium were isolated from organs of some infected plants. Isolation of antagonist microorganisms was carried out from three different Algerian agricultural soils (South, East and north). From a total number of 80 isolated samples in which 18 are bacteria 8 are yeasts and 54 are molds, only 7 strains of the genus *Trichoderma* have presented antifusarial properties. Antifungal activity observed in *Trichoderma* can be due to the production of extracellular enzymes. A range of enzymatic tests has been realized in order to verify this hypothesis. It was about testing chitinase, cellulase, protease, and lipase production. Indeed, all of the isolated species, mentioning : *Trichoderma album, T. virens, T. viride* and *T. longibrachiatum*, are capable of producing chitinase, cellulase, protease, lipase and laccase. These enzymes are responsible of pathogen cellular wall degradation. Several studies have explained the abundance of *Trichoderma* species in ecosystems, by their capacity of producing many bioactive substances and enzymes, they are effectively an important knot in biological chains.

Keywords: Trichoderma, lytical enzymes, biological fight, mycoparasitisme.

# Session 02

# **Health and Environment**

### Keynote 3

# Unraveling the activity and diversity of Ammonia Oxidizing Microorganisms in natural and artificial ecosystems.

#### Luciano Beneduce & Micol Bellucci

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#### Abstract

Nitrification, the two-step oxidation of ammonia to nitrate via nitrite, is a fundamental process of the N-cycle in natural and artificial ecosystems. The first step of nitrification, the ammonia oxidation to nitrite, is carried out by the ammonia-oxidizing microorganisms (AOM), which include Bacteria (AOB) and Archaea (AOA). Water and soil environments, being them natural, influenced by human activities or artificial, are worldwide influenced by nitrification processes, since biological nitrogen fixation and anaerobic nitrogen losses by denitrification are strictly connected by it. In marine environments nitrification process is responsible for about half of the nitrate consumed by growing phytoplankton at the global scale. In soil, AOM influence the global nitrogen transformations and the net balance of plant available nitrogen forms. Yet, in wastewater treatment plants (WWTPs), nitrification is of primary importance for the removal of ammonia, and consequently preventing eutrophication of lakes and rivers. One of the major issues in bioreactor design and management, in which AOM ecology and physiology is essential to the process, is the possibility to correlate efficiency of the process with the taxonomic and functional diversity. Also, for soil environment the diversity of ammonia-oxidizing Bacteria and Archaea and the dynamic of their relative abundance and metabolic activity is a key issue to assess the effect of agricultural input on the quality of crop production and on the maintenance of soil fertility. With the aim of explaining, and controlling, the seasonal failures of the biological nitrogen removal in a wastewater treatment plant (WWTP) treating high saline wastewater (1), we investigated the correlation between the operating conditions and parameters of the plant with the structure and activity of the AOM communities. Though few taxa of alophilic/alotollerant *Nitrosomonas* spp. characterized the ammonia oxidizing bacterial community, a strong correlation between changes in environmental parameters (particularly dissolved oxygen, salinity and temperature) and shifts in the bacterial and AOB communities (assessed by PCR-DGGE and 16S rRNA gene clone libraries analysis) was observed. Nevertheless, the dominant AOB species were always present, independently on the efficiency of the WWTP. The reduction of nitrification performance was therefore linked to the reduction of activity and abundance of the dominant AOB populations without any emergence of AOB better adapted to the changed environmental parameters. The lack of functional redundancy, exacerbated by the AOB slow growth that reduces the rate of recovery following rapid environmental and operational condition changes, was found to be the driving causes of nitrification failure. In WWTPs, nitrification process fails unexpectedly, even in welldesigned and operating systems. The reliability of the system seems to be positively correlated to the microbial diversity therein. With the aim of managing the AOB diversity of nitrifying reactors, we integrated notions of theoretical ecology into the reactor design. The predictions of the resource ratio theory (RRT) developed by Tilman in 1982 were applied to set up and operate nitrifying bioreactors (3). According to the theory, low growing limiting resources, namely oxygen and ammonia inputs, should select for a more diverse AOB community. Hence, reducing oxygen levels, and prospective energy consumption for aeration, might have the simultaneous benefit of reducing the risk of nitrification failure. We successfully manipulated the AOB community structure of lab-scale nitrifying systems according to the two growing limiting resources supplied. Different amounts of oxygen and ammonia selected for AOB communities different in richness and composition as shown by DGGE and clone libraries analyses of AOB 16S rRNA genes. The low concentration of both resources selected for a community with the highest number of species, while the AOB community in the reactor with high oxygen and nitrogen input was the least diverse, in agreement with the predictions of RRT. This suggests that the principles of RRT have the potential to help predicting and possibly managing some characteristics of the microbial communities in WWTPs, in particular, and open microbial

systems, in general. The role of nitrifying microorganisms in soil can be considered negative if excess of ammonium is oxidized to leachable nitrate (anion form) and if increased denitrification to gaseous nitrogen is coupled with nitrification. Addiction of biochar is considered promising for carbon sequestration, soil amendment and has also effect on N-cycle. Biochar amendment of agricultural soil can affect the AOM activity and consequently suppress or promote nitrification, depending on char feedstock, soil characteristics, environmental conditions, agricultural inputs and cropping system. We evaluated the putative impact of biochar addiction on the diversity of AOM in soil cropping systems (3). Our goal was to assess, and possibly correlate, the role of biochar in shaping AOM communities in short term cultural cycles, also considering the interaction with the rhizosphere. The addiction of different char quantities on soil in zucchini crop system did not influence the overall bacterial and archaeal composition during the crop cycle. A strong effect on the overall microbial communities was evidenced within different soil fractions (rhizosphere soil R, soil surrounding root SSR, free soil FS). Only for Archaea the effect of biochar was evidenced between untreated and higher dose treated soil. When focusing on AOM, we found that both AOA and AOB are deeply influenced by the fraction of provenience. Also, the quantity of biochar added did impact the AOB and AOA communities tough with different effects. When considering separately each soil fraction, for AOB the char influence was exerted by mitigation of rhizosphere effect at higher doses. On the other and, if char doses were considered separately, AOA communities showed to be significantly affected only at level of R soil fraction and at high doses treatment.Complex microbial ecology of nitrifying consortia in natural and artificial environments needs to be still fully unraveled and exploited, with the objective of managing nitrogen balance and preventing unwanted gain/losses of nitrogen in a given system. A better understanding of how ammonia oxidizing communities are shaped by-, organized in- and interact with- the environment is a fascinating field of research that has an enormous applicative potential.

# **ORAL COMMUNICATIONS**

# **HE-O1**

# Estimation of incidence of surgical site infections and identification of different types of pathogenic bacteria and evaluation of risk factors in orthopaedic department in Benghazi Medical Center- Libya

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### Abstract

Surgical Site Infections (SSI) are serious complications of operative procedures and are associated with significant morbidity and mortality, the annual cost of SSI to the U.S health care system is more than 1 billion \$. The importance of the SSI estimation has been made a large concern by countries. In order to evaluate SSI in orthopaedic department of Benghazi Medical Center we examined 155 patients that had done surgical operations. The sample from the surgical site of operations is taken from those patients, and microbiological study had been done to surgical site accompanying statistical study of file data to study the risk factors. This study reveals that the incidence of surgical site of infection in orthopaedic department in Benghazi Medical Center is 38.7% and the bacteria that may cause surgical site infections are following: Staphylococcus aureus, Escherichia coli, Pseudomonas as aeruginosa, Staphylococcus epidermidis, Klebseilla spp., Proteus spp., Streptococcus pyogenes, Acinetobacter spp. Moreover the risk factors that may increase of surgical site infections include: Male sex patients, the foreigners, age between 21-30 years of age, operations done on the lower limbs, patients with previous health problems, patients that stayed between one day to one week, patients who operated in cold type operations, the use of drainage and in patients who did not receive antibiotics pre-operative.

**Key words**: Surgical Site Infections, Orthopaedic patients, microbiological study, pathogenic bacteria, risk factors.

# Isolation, Identification and Removal of Microalgae from Keddara Dam Water using Natural Bioflocculant

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# Abstract

Microalgae pose a great danger to the safety of drinking water in the world due to their toxic effects in vertebrates. These microorganisms are generally finely dispersed in water, if they reach high densities, they can increase turbidity and thus cause serious problems in water treatment process. In this study, isolation, identification and removal of persistent microalgae from raw water of Keddara dam has been investigated in order to remedy the presence of microalgae problems. After enrichment in the middle "CONWAY", microscopic observations of algal strains present in the raw water, through an optical inverted microscope magnification "40x" were carried out. It has been noticed a dominance of diatomophycees, Chlorophyceae (Crucigenia, Coelastrum, Scenedesmus, Aphanizozomenon), Cyanobacteria (Oscilatoria) Trebouxiophyceae (Chlorella) classes. As part of the water treatment and environmental protection, a new biodegradable natural product has been tested as an advantageous bioflocculant in terms of coagulation flocculation treatment. A discount rate of 82.5% for turbidity has been obtained with 0.5 mg/l of bioflocculant used as primary coagulant at natural pH (8.04). An improvement of turbidity removal of 93.2% has been reached when the same biofloculant was used as aid coagulant to Alum. Microalgae removal efficiency increased as charge of these micro-organisms increased. Considering the locally availability of this biofloculant and its nontoxic effect, it could be applied to remove microalgae and remediate dam water.

Keywords: Bioflocculant, dam water, identification, Isolation, Microalgae

# Characteristics of extended-spectrum β-lactamase- (ESBL) and AmpC beta-lactamaseproducing Enterobacteriaceae isolated from water samples in Tunisia

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# Abstract

The presence of extended-spectrum  $\beta$ -lactamase and acquired AmpC  $\beta$ -lactamase producing Enterobacteriaceae (ESBL-Eb and AmpC-Eb) was analyzed in 64 wastewater (WW) and 50 surface water (SW) samples in Tunisia. Twenty-five of WW samples (39%) contained ESBL-Eb or AmpC-Eb, but none of SW samples did. ESBL or AmpC genes detected were as follows: bla<sub>CTX-M-1</sub> (10 Escherichia coli), bla<sub>CTX-M-15</sub> (eight E. coli, one Klebsiella pneumoniae, one Citrobacter freundii), bla<sub>CTX-M-14</sub> (one E. coli) and bla<sub>CMY-2</sub> (four E. coli); *bla*<sub>TEM-1</sub>, *bla*<sub>OXA-1</sub> or *bla*<sub>SHV-1</sub> genes were also found in 72% of these isolates. The ISEcp1, orf477 or IS903 sequences were found upstream or downstream of bla<sub>CTX-M</sub> genes. Class 1 integrons were present in 16 of the 25 ESBL-Eb or AmpC-Eb strains (64%), and contained five different gene-cassette arrays. Most of strains (76%) showed a multiresistant phenotype and *qnr* genes were identified in four strains. Molecular typing of ESBL/CMY-2-producing E. *coli* isolates showed 23 different PFGE-patterns and 15 different sequence-types, these strains were ascribed to phylogroups A (10 isolates), B1 (4 isolates), D (6 isolates) and B2 (3 isolates). From 1 to 5 plasmids were detected in each strain (size from 30Kb to > 240Kb) and the ESBL or AmpC genes were transferred by conjugation in 69.5% of E. coli strains. In conclusion, ESBL-Eb and AmpC-Eb strains might be disseminated by WW to other environments, including the human or animal niches, widening the actual problem of global dissemination of multidrug resistance.

# Phenotypic and genotypic patterns of *Salmonella* isolates from human patients in two Algerian regions

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#### Abstract

An epidemiological survey of *Salmonella* from human patients was carried out in two regions of Algeria from 2008 and 2009. During the 2 years, 67 non-duplicate S. enterica isolates recovered from community- and hospital-based patients with acute diarrhoea were collected from all three Annaba hospitals, and routinely analysed at the microbiology laboratory in Dorban hospital. The 67 isolates were subjected to antimicrobial susceptibility profiling using the CLSI guidelines. The genetic diversity was assessed by three subtyping methods; Enterobacterial Repetitive Intergenic Consensus -PCR, Insertion Sequence-PCR, and Pulsed Field Gel Electrophoresis. The genes for antibiotic resistance among the 67 human strains were also performed. The 3 genotyping methods, particularly the PFGE showed the clonality of serotype S. Typhimurium, confirming the diffusion and the persistence of the same clone through the field, and in our regions, but also the polymorphism of serotype Enteritidis indicates the diversity of potential reservoirs of Salmonella. We then identified the genes for antibiotic resistance among the 67 human strains collected during the study period. All of the 50 ESBL-producing Salmonella isolates were positive for the  $bla_{CTX-M}$  gene. Among them, 43 and 35 isolates also harboured the *bla*<sub>CMY</sub> and *bla*<sub>TEM</sub> genes, respectively. Twenty-two out of these 50 ESBL-producing Salmonella isolates showed varied levels of resistance to the aminoglycosides tested (gentamicin, kanamycin), and were screened for detection of the armA methylase gene by PCR. The armA gene was detected in eighteen Salmonella isolates. Sequencing of the *bla*CTX-M genes from the 18 *armA*-positive isolates allowed identification of the CTX-M-15 determinant.

Keywords: Epidemiology, antimicrobial susceptibility patterns, PFGE profiles, resistance genes

# Antibiotic resistance genes and virulence factors of *Escherichia coli* isolated from birds

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#### Abstract

Antibiotic resistance has become a major clinical and public health problem. Indeed, means of transport and bird migration constitute a speed way to disseminate resistance genes acquired from environmental bacteria. Our study aim to investigate the resistance mechanisms developed by *Escherichia coli* isolated from birds faeces collected in northern Tunisia. Biochemical and molecular tools allowed the identification of 51 strains of *E.coli* among 75 of Enterobacteriaceae isolates. The results of standard antibiograms showed strong resistance to amoxicillin (49.01%), ticarcillin(53%) and tetracycline (37.35%). Only imipenem (0%) and gentamicin (2%) were the most active and effective antimicrobial agents. For the other antibiotics tested, resistance varied between 11.76% and 25.49%. Interestingly, 12 E. coli strains exhibited resistance to cefotaxime. In fact, synergy test and molecular study targeting cefotaxime resistance genes revealed that the totality of tested strains were ESBL producing and harbored  $bla_{CTX-M15}$  gene. Moreover, investigated strains presented also the tet(A), qnrB1and *aac*(6')-1b genes responsible for bacterial resistance to tetracycline and ciprofloxacin. We detected also the presence of *int1* gene in 11 strains resistant to trimethoprimsulfamethoxazole. The study of virulence factors highlighted that ESBL-producing strains harbored virulence genes *fimA*, *aer*, *papC* and *eae*, which encodes fimbrial adhesins and pili. Clonality among strains has been studied by pulsed-field gel electrophoresis (PFGE) using the XbaI restriction enzyme. This study proves that birds constitute an important reservoir of antibiotic resistant E. coli. This phenomenon of bacterial resistance is becoming a global threat, emphasizing the need for the application of hygiene measures, rational prescription of antibiotics and the establishment of a system to monitor the evolution of epidemiological strains.

# Antimicrobial Resistance and virulence genes of *Staphylococcus aureus* from Bovine mastitis milk samples in Tunisia

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#### Abstract

Staphylococcus aureus is a main cause of bovine mastitis and a major pathogen affecting human health. The spread of methicillin-resistant Staphylococcus aureus (MRSA) has become a significant concern for both animal health and public health. The prevalence of S. aureus and the emergence of MRSA/MSSA from mastitis is currently unknown in Tunisia. A total of 212 Milk simple from mastitis bovine with clinical symptom were obtained for a period 2013-2014 from different big farms in Tunisia. Swabs were subcultured on Baird Parker (BP) and Orasb medium (oxacillin Resistance screening Agar Base oxford) for S. aureus methicillin-resistant (MRSA). S. aureus was distinguished from coagulase negative Staphylococci by the coagulase test and was then confirmed by amplification of the nuc gene. Respectively, MRSA was detected in 3 of these 212 samples (1, 41%). The mecA gene was detected in 3 S. aureus, additionally, 7different sequence-types (STs) were detected by MLST, ST97 (9 isolates), ST5 (1 isolate), and five of them news. Most of MRSA showed susceptibility to the tested antibiotics with exceptions: 1 strain carrying blaZ gene and 1 strain carrying msrA gene. Methicillin-susceptible S. aureus (MSSA) were recovered in 12 of the 212 samples (5.66%), Most of MSSA showed susceptibility to the tested antibiotics with exceptions: (penicillin 2 strains carrying *blaZ* gene), (erythromycin 1strains carrying *ermC* gene). All isolates strains were negative for Panton-Valentine leucocidin(PVL), exfoliating EtA and EtB, Thestaphylokinase (sak), Enterotoxin A (sea), and the (scn) gene was detected in one S. aureus methicilline susceptible isolate and The Toxin shock toxic (TST) was detected in 4 strains S. aureus, by PCR. Moreover, Enterotoxin P (sep) and chp genes were not found. The presence of S. aureus methicillin resistant and susceptible emphasizes the importance of identification of staphylococcus when an intramammary infection is present because of the potential risk of horizontal transfer of resistant genes and virulence gene among staphylococcal species.

### Use of biochemical markers in the early detection of bacterial meningitis

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#### Abstract

Bacterial meningitis is related to the invasion of the cerebrospinal fluid (CSF) by bacteria that develops. The number of cases of this bacterial meningitis is estimated at over a million a year worldwide. Bacterial meningitis in children are in all cases, a therapeutic emergency involving early suspicion of a diagnosis must be confirmed by the CSF after lumbar puncture. Our goal is to establish the relationship between the rate of CSF glucose and bacterial presence in CSF to facilitate the confirmation of early diagnosis and provide adequate and immediate antibiotic therapy. Twelve CSF samples obtained from hospitalized children with clinical symptoms (fever, convulsions ...) were analyzed in order to assay CSF protein concentration and CSF glucose by using enzymatic methods; and glucose. After, analyzes were performed on the blood and CSF were obtained the following results: - Biochemical CSF shown, moderate protein level and a variable CSF / blood glucose ratio of a child for another 17% (<0.40) and 83% (> 0.40). These results indicate that normal CSF glucose does not exclude the diagnosis of this type of meningitis, and it could not be a reliable marker in the confirmation of a bacterial infection.

Keywords: Bacterial meningitis; Cerebrospinal fluid; Children; CSF glucose.

# First report of Extended Spectrum β-lactamases among clinical isolates of *Klebsiella pneumoniae* in Gaza Strip, Palestine

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#### Abstract

The objective of this study was to assess the occurrence of extended-spectrum  $\beta$ -lactamases (ESBLs) in *Klebsiella pneumoniae* isolates from clinical samples in Gaza hospitals, Palestine in 2013 and to characterize ESBLs, their genetic environments and the associated resistance genes. Sixteen broad-spectrum-cephalosporin-resistant K. pneumoniae isolates were recovered between April and June 2013 in Palestinian hospitals, Gaza Strip. The characterization of ESBL genes and their genetic environments, detection of associated resistance genes and the presence and characterization of integrons were performed by polymerase chain reaction (PCR) and sequencing. The following ESBL genes were identified:  $bla_{\text{CTX-M-15}}$  (13 isolates),  $bla_{\text{CTX-M-14}}$  (2 isolates) and  $bla_{\text{CTX-M-3}}$  (1 isolate).  $bla_{\text{SHV}}$  -type represented 75% of the strains. The main enzyme present was  $bla_{SHV-1}$ , found in 7, the second enzyme was  $bla_{SHV-5}$ , detected in 3 isolates and we found One isolate of  $bla_{SHV-12}$  and  $bla_{SHV-12}$ 33. The *bla*<sub>TEM-1</sub> gene was detected in 7 *bla*<sub>CTX-M</sub> producing strains. Of the isolates investigated, 3 isolates were found to produce  $bla_{OXA-1}$  and associated with  $bla_{CTX-M}$  and *bla*<sub>SHV</sub>. The *ISEcp1* sequence was found upstream of *bla*<sub>CTX-M</sub> genes in all strains (in four cases truncated by IS26). The orf477 sequence was detected downstream of bla<sub>CTX-M-1</sub> group in all thirteen  $bla_{\text{CTX-M-15}}$  strains and in the unique  $bla_{\text{CTX-M-3}}$  harboring stain. The IS903 was detected downstream of the two strain of *bla*<sub>CTX-M-14</sub> gene. Seven of the strains carried class 1 integrons and gene cassette arrangements detected were, dfrA5-aadA4 (three isolates), dfrA12-aadA2 (two isolates), dfrA17-aadA5 (one isolate) and aadA1 (one isolate). The ESBLs are detected in K. pneumoniae of clinical origin in Gaza, being the first time that this mechanism has been detected in Gaza strip hospitals.

Key Words: K. pneumoniae; ESBL; bla<sub>CTX-M-15</sub>; Gaza strip; Palestine

#### Toxicity of ammonium based ionic liquids towards "Shewanella sp."

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#### Abstract

Room-temperature ionic liquids (ILs) are a class of novel green chemicals being designed to replace traditional volatile organic solvents in industrial processes but these new solvents can be more toxic towards cells than conventional solvents; this must be taken into consideration with regard to their fate and persistence in the environment. There is a relationship between the structure of the cation and the anti-microbial activities of the ionic liquids. Considering that no general influence of the anionic compound in the ionic liquids on toxicity could be found, although they seem to modulate toxicity in some cases. In this study we want to investigate the relationship between the structure of the cation and the anti-microbial activities of ammonium based ionic liquids towards the marine bacteria "*Shewanella sp.*. The bacterial growth was inhibited progressively as the length of the 1-alkyl chain of the cation side was increased; it was found also that the toxicity of ammonium ionic liquids can be due to the anionic moiety.

Keywords: ammonium, ionic liquids, alkyl chain lent, toxicity, Shewanella sp.

# POSTERS

# Antibiotic resistance determinants of multidrug-resistant *Pseudomonas aeruginosa* clinical isolates in eastern Algeria

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# Abstract

*Pseudomonas aeruginosa* is considered to be a major opportunistic pathogen contributing to the installation of nosocomial infections in immunocompromised individuals. *Pseudomonas* infections are many, among them respiratory, urinary tract infections and wound infections. Factors that increase the pathogenicity of *Pseudomonas aeruginosa* in the inpatient environment are: growth in the form of biofilm and multidrug resistance of this germ to antibiotics. The study of sensitivity to antibiotics was conducted by the method of diffusion agar according to the recommendations of the Committe of the French society of Microbiology (CASFM 2011). Antibiotic susceptibility test have shown that the majority of the strains forming biofilms are multi resistants with a percentage of 57.90%. The strains involved in this study have a very high resistance, the presence of strains multiresistant is a problem in hospital mostly in intensive care unit where patients are inmostly intubated, and their immune systems are unable to cope with the bacteria multi resistant.

Key words: Pseudomonas aeruginosa-multidrug-resistant bacteria-biofilm formation.

# Identification and characterization of lactic acid bacteria during spontaneous fermentation of tomato fruits

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#### Abstract

Lactic acid bacteria (LAB) are a group of gram-positive, cocci or rods, non-spore forming bacteria, which produce lactic acid as the major product during the fermentation of carbohydrates. They are commonly isolated from fermented foods and dairy products. LAB found different industrial application regarding their particular physiological and biochemical characteristics such as aromatic compounds, exopolysaccharides and organic acids The present work aimed identify LAB during spontaneous production. to fermentation of tomatoes fruits and to assess their biotechnological properties through the evaluation of aromatic compound and exopolysaccharides production, acidifying power and antagonistic activity against several bacterial species. Using the variability of the intergenic spacer 16S-23S and 16S rRNA gene sequences, Lactobacillus and Enterococcus were the most encountered genera. All the LAB retrieved strains were able to produce exopolysaccharides and aromatic compounds. While, only 16% of the investigated strains, presented rapid acidification of the medium. Antibacterial activity of LAB was evaluated against Pseudomonas viridifla, Pseudomonas poae, Bacillus pumilus and Staphylococus homonis. The highest inhibitory activity was recorded for Lactobacillus plantarum strains against Staphylococus homonis. The promising results of this investigation underline the importance of LAB as a starter player for fermented food products and industrial application. Further experiments will be conducted to assess microbiological and organoleptic properties of fermented tomatoes using selected LAB based-starters.

**Keywords**: Lactic acid bacteria, aromatic compounds, exopolosaccharides, acidifying power and antibacterial activity.

# Molecular characterization and evaluation of inhibitory potential of Lactobacilli strains isolated from mother's milk against uropathogenic microorganisms

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#### Abstract

The vagina is a complex habitat for micro-organisms. 3/4 of the women suffer at least once in their life from vaginal mycosis and 1/3 are concerned with bacterial infections of the vagina (.The aim of our work was to characterize and to select the probiotic Lactobacilli isolates as probiotic candidates for the bacterial vaginitis and the vaginal mycosis therapy. Fourteen strains of Lactobacillus sp. were isolated from the mother's milk of Algerian women. The antibacterial activity of Lactobacilli isolates against uropathogenic microorganisms (Candida albicans L2 and Escherichia coli SI1) was studied by the well diffusion method. The nature of the inhibiting molecules was given according to the semi quantitative method of production of hydrogen peroxide, the study of the acidifying activity as well as the concentration of proteins for the detection of bacteriocins. The results obtained show a very strong activity (diameter of inhibition between 20 and 30 mm) at two strains of Lactobacilli (Lactobacillus rhamnosus LB16 and Lactobacillus rhamnosus LB50). These inhibiting activities are due mainly to the acidity and hydrogen peroxide. The phenotypical identification of the two selected strains was confirmed by the sequencing of the genes 16S rDNA. Other work by application in vivo of the tests realized in vitro, as well as the study of their capacity of adhesion will be necessary to better apply the concept probiotic at the urogenital level.

Key words: Lactobacilli, characterization, antimicrobial activity, vaginal infections.

#### Antimicrobial activities of synthetic thio- and dithio-carbamates

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#### Abstract

During the last decade, the emergence of microbial resistance to the antibiotics poses a serious concern for medical professionals. Thus, the research for new molecules with promising antimicrobial ability is needed. In this study, synthetic molecules of thio- and dithio-carbamates with the formula (R-HN-C(O)-X-R') were tested for their activity against nine strains of Gram positive and Gram negative bacteria (Enterococcus faecalis ATCC 29212, Enterococcus faecium ATCC 19436, Staphylococcus aureus ATCC 25923, Staphylococcus aureus ATCC 6538, Staphylococcus aureus CIP 4.83, Bacillus cereus 49, Escherichia coli DH5a, Escherichia coli ATCC 8739 Pseudomonas aeruginosa and Salmonella sp) and two fungal strains (Aspergillus niger, Penicilluim sp). The results showed that fungal strains were more sensitive than bacterial ones. In addition, fluorinated compounds were found to be more potent when bearing an X = S and an R= ethyl group compared to X =O and R = phenyl or peridyl groups especially against Gram positive bacteria. All the tested compounds showed good activity against fungal strains with MIC values ranging from 7.8 µg/ml to 62.5 µg/ml. However only fluorinated dithiocarbamates showed good antibacterial effect with MIC values of 3.90 µg/ml against Bacillus cereus 49, 7.81 µg/ml against Pseudomonas aeruginosa and 15.62 µg/ml against Staphylococcus aureus 6538.

Keywords: Thiocarbamates, dithiocarbamates, antibacterial and antifungal activities.

# Fungal isolates and their toxigenic potential from algerian mussels and their immediate environment

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#### Abstract

The littoral represents the final compartment of the entropic rejections generating great quantities of organic matters. These areas correspond to an ideal place for the development of saprotrophic fungal communities. It is well established that micromycetes may occur in marine environment. Indeed, their presence has been reported from sediment, water and shellfish samples. These reports suggest that marine environment could represent genuine saprotrophic fungal tanks. Toxigenic saprophytic fungi were isolated from samples of Mussels, sediment and seawater along the Algerian Western Coastal Areas. More than 250 strains belonging to 12 different genera had been isolated with a clear predominance (85%) of *Penicillium, Aspergillus,* Muccorales, *Cladosporium, Trichoderma* and *Fusarium.* To assess the risk of poisoning due to the presence of these fungi in shellfish farming areas, the strains were cultured in liquid medium, filtered and extracted. Brine shrimp bioassay was applied for screening toxic strains. A third of the tested fungal strains proved to be toxic, thus showing a considerable toxigenic potential of the micromycetes isolated in these study area.

Key words: Micromycetes, brine shrimp bioassay, Toxigenic potential, Western Algerian littoral.

# Ticks infesting brown hare (*Lepus capensis*) and wild rabbit (*Oryctolagus cuniculus*), as potential risk for transmitting pathogens to humans in center of Algeria

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### Abstract

The wild rabbit and the hare are lagomorphs which constitute a small game very appreciated by the Algerian hunters. They represent an alternative to the development of the ecotourism. This study's objective was to identify ticks infesting hares and wild rabbit in the wild center's of Algeria to estimate potential risk of agents' pathogens that can be transmitted by these parasites. Between July, 2014 and March, 2015, 62 hares and wild rabbits were examined for ticks and identified them based on their morphologic characteristics, 3 genus of ticks are identified as 130 Ixodes spp, 77 Rhipicephalus spp and 34 Hyalomma spp. 4 species, Ixodes ricinus which is commonly found in Algeria and can transmit Borrelia burgdorferi, Francisella tularensis, Ehrlichia spp, Rickettsia spp, Coxiella burnetii, A. phagocytophilum, Candidatus N. mikurensis, virus and parasites like Babesia divergens and Babesia. Venatorum, Rhipicephalus sanguineus as principal vector of Rickettsia conorii and it can transmit Leishmania spp .Rhipicephalus pusillus, in the past; it was considered as reservoir of Spotty fever. In addition, it can be vector of Rickettsia sibirica, Rickettsia massiliae and Coxiella burnetii. Hyalomma marginatum as principal vector of hemorrhagic crimen Congo virus and Dhori virus. Studies to assess whether these ticks could limit the survival and fitness of wild leporidae and affect their conservation status are needed. Moreover, it is necessary to investigate whether these ticks are infected with pathogens of medical and veterinary concern.

Key words: Brown hare, Wild rabbit, Ticks borne diseases, Ixodes, Rhipicephalus, Hyalomma

# Adhesion and biofilm formation by *Legionella pneumophila* and *Pseudomonas aeroginosa* in water distribution systems: role of supports and temperatures

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#### Abstract

Legionella pneumophila, the aetiological agent of 90% of legionellosis cases, is a Gramnegative bacterium hydrotellurique originally found in both natural environments (lakes, rivers) than in artificial environments (cooling towers, hot water networks ...), where it is able to form biofilms. These biofilms represent a sophisticated network of metabolic and architectural interactions, which concentrates nutrients and protects the microbial residents from physical, chemical and biological hazards. Legionella pneumophila with the ability to join and form biofilms in water distribution systems depending of environmental conditions. The materials for water distribution systems vary according to their ability to foster growth and biofilm formation by L.pneumophila. The aim of our study was to determine the kinetics of adhesion and biofilm formation by Pseudomonas aeruginosa and L.pneumophila on polystyrene and five different surfaces (galvanized steel, Polypropylene, PVC and copper) commonly used in systems distribution of water three growth temperatures 20°C, 37°C and 44°C. L. pneumophila serogroup 2-15 and *Pseudomonas aeruginosa* showed a greater capacity than serogroup 1 L.pneumophila to adhere and form biofilm on the majority of materials tested at 20°C to 37°C and 44°C. Only the copper inhibits the growth of biofilm and colonization of water systems by the stem 3 at all tested temperatures. Understanding these mechanisms is part of the purpose of the risk control contamination of these surfaces and material selection of the appropriate conduct can minimize the possibility of biofilm development partner in water distribution systems and limitation of infections with these bacteria causing Legionnaires.

Key words: Legionella pneumophila, Pseudomonas aeruginosa, adhesion, biofilm, supports.

# High level of biofilm production and antibiotic resistance rates virulence factors and phylogenetic groups in *Escherichia coli* isolates from Environment in Tunisia

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#### Abstract

Objectives of our work were to determine the frequencies of antibiotic resistance in a collection of *E. coli* isolates recovered from various environment origins, the study of phylogenetic groups, as well as the determination of classes of integrons carrying the genes for antibiotic resistance and gene cassettes involved. We found a high percentage of resistance for the following antibiotics: tetracycline, trimethoprim-sulfamethoxazole, sulfonamides, streptomycin, nalidixic acid and ampicillin. Integrons of class 1 and 2 have been identified in the majority of our isolates. Conserved region, *qac-sul1* of the integron class 1 was detected in isolates with class 1 integron. Studied isolates belong mainly to phylogenetic groups A and B1 (70%) and are considered to be of low pathogenicity. Groups B2 and D, most pathogens, represent 30% of the collection studied. By the congo red agar method (CRA) isolates were positive for *in vitro* biofilm production. By the crystal violet (CV) method, (40%) strains were classified as strong positive, and (7.5%) were no biofilm producers. Similarly, the tetrazolium salt essay (MTT) method showed that (92.5%) strains were classified as highly strong positive, (6.3%) moderate positive and one (1.2%) weakly positive. While in tetrazolium hydroxid (XTT) method, all the strains were biofilm producers.

Keywords: Escherichia coli, antibiotic resistance, the integron, phylogenetic groups, biofilm.

### Comparative evaluation of matrix-assisted laser desorption ionisation-time of flight mass spectrometry (MALDI-TOF) and conventional phenotypic-based methods for identification of *Serratia marcescens*

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#### Abstract

Conventional phenotypic-based methods used for bacterial identification relies on culturebased methodologies requiring 24 h for isolation and an additional 24 to 48 h for species identification. Matrix-assisted laser desorption/ionization time of flight mass spectrometry (MALDI-TOF MS) has emerged as a potential tool for microbial identification and diagnosis. During the MALDI-TOF MS process, microbes are identified using either intact cells or cell extracts. The process is rapid, sensitive, and economical in terms of both labor and costs involved. In this study, fifty-five Serratia marcescens isolated from urine samples have been included. Identification of bacterial colonies was performed under routine clinical laboratory conditions based on morphological and biochemical criteria. While using MALDI-TOF-MS, the sample is mixed with the matrix placed on a metal plate, the target. A laser source is directed on the target to ionize the sample molecules. The ions are then detected by measuring the time it takes the individual particles to reach the detector. The speed of each particle depends on the mass / charge ratio. Larger molecules will take longer to reach the detector, while the smaller molecules arrive faster. Once arrived at the ion detector, the signal is amplified and sent to a computer which processes the data and provides the results in the form of spectrum. Our results show that phenotypic methods used were not enough to confirm the real species of the genus Serratia. However, the MALDI-TOF MS confirm the identification of Serratia species and show that the 55 are S. marcescens with good score value between 1.910 -2.302.

Keywords: Serratia marcescens, MALDI-TOF MS, Phenotypic identification,

# Study of Cytomegalovirus infection and characteristics of CMV in Tunisian ulcerative colitis patients

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#### Abstract

CMV colitis is a common manifestation of CMV end-organ disease. CMV which usually remains asymptomatic in the healthy adults can cause a symptomatic disease in the immunocompromised patients. The risk of infection with Cytomegalovirus increases in ulcerative colitis (UC) patients as a result of receiving immunosuppressive agents. This study aims to evaluate cytomegalovirus (CMV) infection in Tunisian ulcerative colitis (UC) patients and to analyse CMV genes UL55 and UL138 in peripheral blood leukocytes (PBLs) by polymerase chain reaction (PCR). A total of 25 patients with ulcerative colitis (UC) were included in this study.30 Non-CMV colitis patients who were age- and sex-matched were selected as controls for each case. CMV-specific serum IgG and IgM antibodies were investigated by enzyme-linked immunosorbent assay. CMV genes UL55 and UL138 were detected in PBLs by polymerase chain reaction (PCR), Data indicated that the frequency of positive IgG and IgM anti-CMV antibodies was not significantly different in UC patients and controls. However, compared to the healthy controls, the titers of IgG anti-CMV antibodies in UC patients were significantly higher. CMV infection can be related to the development of UC patients especially, some clinical strains of CMV.

# Microbiological risk evaluation related to consumption of Döner Kebab in Tlemcen, Algeria

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#### Abstract

The objective of this study was to define the microbiological quality of döner kebab consumed in the town of Tlemcen and determine the antibiotic resistance of isolated strains from of the product. Therefore, 20 samples were analyzed, the microbiological evaluation focused on the enumeration of total mesophilic flora, coliforms, *Escherichia coli*, psychrotrophic bacteria, *Staphylococcus aureus* and sulphite-reducing clostridia as well as the detection of Salmonella, Campylobacter, yeasts and molds. 200 isolates were obtained, which 54 identified belonging to three major families: Enterobacteriaceae, Micrococcaceae and Campylobacteriaceae that were able to be identified through phenotypic and biochemical tests. According to the results of the microbiological analysis, Sector 1 revealed the highest contamination rate in FMAT ( $6,7.10^6$  bacteria/g), followed by sector 4. The results of antibiotic resistance display a high resistance to various antibiotics widely used in human and veterinary medicine, except gentamicin, ciprofloxacin and norfloxacin for which no resistance was observed.

Key words: Döner kebab, meat, microbiological quality, antibiotic resistance, Tlemcen.

# Development of an environmentally friendly priming emulsion

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#### Abstract

New granular pavements are usually primed or primer sealed before further surfacing is carried out. The priming process binds the surface layer and provides a water resistant surface suitable for effective bonding to a bituminous surfacing such as a chip seal or asphalt overlay. Conventional primes based on hydrocarbon solvents are subject to several limitations. In particular, under cool conditions the primed pavement is at risk from heavy rain falling soon after priming. This can result in washout of the prime and subsequent entry of contaminated run-off into waterways resulting in environmental pollution. A bituminous emulsion prime was developed to overcome a number of the shortcomings of conventional kerosene based systems including the washout risk, without compromising penetration and pavement binding properties. The work presented extends that initially carried out in 1998 and examines leaching and tensile strength properties of primed laboratory compacted crushed rock samples. Field performance supports laboratory observations.

Keywords: Primer, Prime coat, emulsion, environment.

### Isolation of actinomycetes bacteria from the rhizosphere of the date palm of some desert regions of South East Algeria and study their roles in biodegradation of Phoenix insecticide

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#### Abstract

The different pesticides used in agriculture constitute a significant threat to the environment. Phoenix Lambda Cyhalothrin is a chemical compound from the class of Pyrethroid, Widely used as a non-systemic insecticide for the fight against a wide variety of diseases transmitted by insects. This insecticide is used for several years in Algeria. It is able at high concentrations to cause considerable toxicity to soil and groundwater waters. The urgency of the use of different techniques of dépollutions are more than necessary. Among these techniques, biological processes involving micro organisms are most advantageous. In this study, we are oriented to actinomycetes which are known by their biodegradation capacity of the most varied polymers. Thus, 148 actinomycetes were isolated from desert soil samples of the rhizosphere of the date palm of Biskra, Ghardaïa, Touggourt, 'El-oued and Hassi messaoud regions. These bacteria were collected from three selective isolation medium: The starch casein medium, GLM and glycerol arginine agar medium. All these actinomycetes strains were tested for their ability to degrade a chemical compound, taken as a sole source of carbon and energy. The results indicate that among the 148 isolates, 13 actinomycetes are able to degrade the insecticide. The culture conditions of these bacteria were determined. Our strains are able to degrade a concentration of 500 mg / 1 of Phoenix at a temperature of 30 ° C for 21 days of incubation. This study showed that actinomycetes recovered from the Algerian desert soils, could be an excellent microbial agents used in the bioremediation of contaminated environments with pesticides.

Keywords: Actinomycetes, degradation, Phoenix, bioremediation.

# Seroepidemiological study of infection with *Leptospira interrogans* serovar Hardjo in cattle farms of the Algiers region

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#### Abstract

Leptospirosis is a worldwide distribution zoonosis caused by Leptospira interrogans responsible for heavy loss both economically and on human health and animal. In this work, a seroepidemiological study on the infection by L. interrogans was conducted in cattle farms of the area of Algiers, from March 2013 to January 2014. The total of 19 farms randomly selected were investigated and 205 cattle serum was collected and tested on the one hand by the MAT test to determine the prevalence of specific antibodies 5 serovars of L. interrogans using a 50% agglutination at a dilution  $\geq$  1: 100 as threshold positivite. 29/205 (14.14%) animals were positive to one or more serovars. Leptospira serovars most prevalent were Hardjo and Pomona with 13 samples positive for each (6.34%) followed by Canicola (4.39%) and Grippotyphosa (3.90 %). The less prevalent serovar was Icterohaemorrhagiae with only two positive samples (0.97 %). On the other hand, the seroprevalence of L. interrogans serovar Hardjo was evaluated by the ELISA. A 5 % was obtained by ELISA PrioCHeck. The comparison between different tests used showed that the MAT remains the tool serologic of reference in the diagnosis of an infection with L interrogans. The study of various potential risk factors showed no statistical association with infection with L. interrogans servar Hardjo.

**Keywords :** Leptospirosis, PrioCHeck ELISA, *Leptospira interrogans* serovar Hardjo, prevalence, Algiers.

# Isolation and efficacy of entomopathogenic fungus *Metarhizium anisopliae* for the control of *Culex pipiens* larvae vectors of diseases in Algeria

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# Abstract

In the search for alternative methods to chemical control, the use of the biopesticides is developed more and more against mosquitoes larvae of *Culex pipiens* vectors of infectious diseases. The strains of fungus are most promising from this new point of view. In this study, we did a biological test of the local entomopathogenic fungi *Metarhizium anisopliae* isolated from agricultural soils of Constantine. The susceptibility of *Culex pipiens* larvae to the entomopathogenic fungi *Metarhizium anisopliae* was examined under laboratory conditions. Five doses were used D1=  $10^5$  spores/ml, D2=  $10^6$  spores/ml, D3=  $10^7$  spores/ml, D4=  $10^8$  spores/ml and D5=  $10^9$  spores/ml. The mortality values were then subjected to probit analysis. The larval mortalities were observed for a period of 10 days, LC<sub>50</sub> and LC<sub>90</sub> values were calculated. Results obtained demonstrate that larvae mortality increased with increasing concentration and time of exposure to treatment. The results indicate that *Metarhizium anisopliae* has the potential to be a biocontrol agent for mosquito and is suitable candidate for further research and development.

Key words: Entomopathogenic fungi, Metarhizium anisopliae, Culex pipiens, Biocontrol, Mosquito

# Antagonistic activity of lactic acid bacteria isolated from camel milk of south east Algeria

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### Abstract

Lactic acid bacteria play an important role in biopreservation of foods and intestinal prophylaxis. These bacteria not only produce several antimicrobials during carbon source metabolism, they also compete with other species by acidifying their environment and by rapidly depleting the nutrients. For this, 48 LAB isolated from camel milk were screened for antagonistic activity against five spoilage and pathogenic bacteria which are : Escherichia coli ATCC 25922, Staphylococcus aureus ATCC 25923, Salmonella sp., Bacillus subtilis and Enterobacter cloacae using an agar spot test. An overnight culture of LAB was spotted onto the surface of an MRS plate and incubated in anaerobic conditions for 24h at 30°C to allow the colonies to develop. Approximately 10<sup>6</sup> cfu/ml of indicator strain were inoculated into 10 ml of soft nutrient agar and poured over the plate on which the producer was grown. After aerobic incubation during 24h at 37°C, zones of inhibition around the spots were measured in millimeters. Results show that all strains of LAB tested for their antagonism effect inhibited the growth of the five undesirable bacteria. The inhibitory effect differs significantly from one isolate to another with a minimum diameter equal to 7 mm obtained by the bacterium B5 (Leuconostoc sp.) against S. aureus, and a maximum diameter of 32 mm obtained by the strain O8 (Lactobacillus sp.) against E. coli. From these results, it seems that lactic microflora participates also in health properties and the long preservation period characterizing camel milk.

Key words: lactic acid bacteria, antimicrobial activity, camel milk, spot on the lawn test

# Antimicrobial activity and tlc analysis of bioactive substance extracts from cyanobacteria *Arthrospira platensis*

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#### Abstract

Blue green algae Spirulina platensis it has been used in various ailments and as health tonic The search for cyanobacteria with antimicrobial activity has gained importance in recent years due to growing worldwide concern about alarming increase in the rate of infection by antibiotic-resistant microorganisms. Various strains of Cyanobacteria are known to produce intracellular and extracellular metabolites with diverse biological activities such as antibacterial. To understand the mechanisms of pharmacological actions, the aim of study reported here was to investigate the antibacterial activity, and thin layer chromatographic separation of organic and aqueous extracts of Spirulina platensis were screened. methanol extract and aqueous extract were tested in vitro against species of human pathogenic bacteria; one Gram-positive bacteria (*Enterococcus feacalis*) by the agar-solid diffusion method. The extracts were analyzed by TLC. Eluent: Methanol, chloroforme, ethyl acetate by different concentration and systems (seven systems) previously left to equilibrate for at least 30 min. The chromatograms were evaluated in UV light at  $\lambda$ =366 nm .000 The antimicrobial activity of extracts showed that greater inhibition zone against gram positive bacteria in aqueous extract, than methanol extract .Nevertheless, , methanol extract showed moderate activity. A R<sub>F</sub> value of flavonoïds and phénolic identified in the methanolic extract by nine mobile phases, and two by aqueous extracts by different systems. The preliminary studies on spirulina extracts exhibited their antimicrobial potential which could be exploited further as future antimicrobials for pharmaceutical treatment, natural therapies, food preservation and cosmetic applications.

Key words: Antibacterial activity, *Spirulina platensis, Enterococcus feacalis,* TLC, UV, RF value.

#### Anti- Helicobacter pylori and urease inhibitory effects of polyphenolic extract of Achillea odorata leaves

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#### Abstract

Since its discovery, *Helicobacter pylori* has been increasingly emerging as a major health problem worldwide. In particular, it has been regarded as a major cause of chronic gastritis and peptic ulcer. Due to the emergence of antibiotic resistance among clinical strains of *H. pylori*, new antimicrobials are needed. Methanolic extract of Algerian originated *Achillea odorata* and its content of phenolics and flavonoids were evaluated for the *in vitro* antimicrobial activity against *H. pylori* in addition to its associated urease inhibition. The antimicrobial activity and determination of the MIC of this extract against control strain of *H. pylori was* performed using standard agar diffusion method. Preliminary screening based on specific tests concluded in the presence of substances with large therapeutic values. The total phenolic content confirmed the presence of total soluble phenolics in the extract. In addition, the herbal extract exhibited a strong anti-*H. pylori* activity (MIC 3 mg/ml). For the urease activity, a high activity and wide range was found for *A. odorata* (00-10% at concentrations 125-250 mg/ml). Our results support the tendency to use this extract as an alternative treatment of *H. pylori* infections.

Key words: H. pylori, Achillea odorata, polyphenols, urease inhibition

# Comparison of microbiological quality of seven dairy products originating from Tunisia

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#### Abstract

The current study was carried out to evaluate the microbiological quality of seven dairy products (raw milk, pasteurized milk, UHT milk, fresh cream, butter cream, butter and cream of fresh cream) originating from Tunisia. Nine samples of each product were used to determine the total mesophilic germs, thermophilic, total and fecal coliforms, and, the presence of certain pathogenic bacteria. Indeed, the obtained results show that the microbial load of various germs depends on the type of analyzed product. In fact, raw milk contains the most number of these germs followed by pasteurized milk, butter cream and cream. UHT milk has a null charge in theses germ, indicating the efficacy of UHT treatment. For the prevalence of pathogenic bacteria, we noticed that the microbiological quality of all products is unsatisfactory, except for UHT milk, compared with the standards. For example, the number of *S. aureus* is between 0log CFU / ml (UHT milk, butter) and 5log CFU /ml (raw milk). While *E. coli* and *L. monocytogenes* are present in the majority of products except in the UHT milk and fresh cream. The absence of *Salmonella* in the majority of products tested except in the case of raw milk. Therefore, a potential risk of foodborne illness may be implicated in some dairy products.

# Assessment of sanitary quality of some foods of animal origin sold in Djelfa city (Algeria)

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#### Abstract

This study was conducted in order to assess the bacteriological quality of some foods of animal origin, commercialized in local markets in Djelfa city. A total of 109 samples, divided between 30 raw milk, 30 traditional cheese, 30 eggs and 19 chickens, were purchased as sold to consumers and analyzed. Bacterialogical analysis were based on enumeration of (TBC andTtC) as flora of hygienic interest, *Staphylococcus aureus, E. coli, Salmonella*, Sulfite reducing Clostrida and *Brucella* antibodies (realized by the Ring-test) as flora of sanitary interest. The results show that hygienic quality was generally insufficient. The log mean total bacteria counts per ml was estimated at 7,16 cfu.ml<sup>-1</sup> for raw milk and 6,6 cfu.ml<sup>-1</sup> for chickens. The average contamination of raw milk by thermotolerant coliforms was 4,79 cfu.ml<sup>-1</sup>, against 4,97 cfu.ml<sup>-1</sup> and 4,42 cfu.ml<sup>-1</sup> respectively for cheese and chickens. *Staphylococcus aureus* was detected in 25,31% while sulphite reducing clostridia was present in milk with 16,66%. In addition, 18,34% of samples were positive for *E. coli. Salmonella* was isolated in 5 samples (2 eggs and 3 chickens) with a prevalence of 4,58%. Paradoxically, samples of milk and cheese were free from *Brucella* antibodies.

# Occurrence and distribution of multiple antibiotic-resistant bacteria of Enterobacteriaceae family in waters of several wadis from Algeria

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#### Abstract

Antibiotic-resistant organisms enter into water environments from human and animal sources. These bacteria are able to spread their genes into water-indigenous microbes, which also contain resistance genes (Baquero et al., 2008). This study deals with antibiotic resistance profiles in Enterobacteriaceae family isolated from twenty two wadis situated in North of Algeria. A total of 22 enteric bacterial are isolated. Among the isolates, 10 (45.45%) are Escherichia coli, 3 (13.63.1%) Serratia fonticola, 2 (9.09%) Klebseilla pneumoniae pneumonia, 2 (9.09%) Enterobacter cloacae, 2 (9.09%) Klebsiella oxytoca, 1 (4.55%) Enterobacter sakazakii, 1 (4.55%) Proteus mirabilis and 1 (4.55%) Serratia liquificiens. Twenty two antibiotics were used for determination of antibiotic resistance profiles of the isolates. Antibiotic resistance of bacteria was determined by the disc diffusion method. All Enterobacteriaceae isolates exhibited resistant to β-lactams antibiotics especially the carbapenem (Imipenem) (92.50%) and most other classes of antibiotics. In total, 100% of the isolates exhibited multidrug resistance character and all the isolates had a very high multiple antibiotic-resistance index, suggesting the origin of the isolates to be of high antibiotic usage. However, multiple antibiotic resistance (MAR) from all sites could be detected which is possibly due to sewage discharge and input from other anthropogenic sources. This study is only preliminary; extensive research into the possible long-term risks of water borne diseases via the ingestion of these drug resistant bacteria is advocated.

Key words: Antibiotic resistance profiles, Enterobacteriaceae, wadi, MAR, Pollution, Algeria

#### Microbiological quality of marine waters in the region of Skikda – Algeria

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#### Abstract

The bacteriological results obtained in our study on an evaluation of the pollution of the coastal zone of Skikda using sampling of surface marine waters, have enabled us to highlight a high microbiol load far exceeding the limit values laid down by the legislation in force (Algerian). These sometimes major pollution caused by wild discharges of untreated wastewater and its direct consequence on the coastal ecosystem and public health, give to the existence a character of insecurity and fragility.

Keywords : pollution, coastal, Algeria, coliforms, fecal streptococci.

# Antibacterial activity of endophytic fungi isolated from *Urtica dioica* leaves (nettle common)

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#### Abstract

Scientists have recently focused their interest on the endophytic fungi that colonize medicinal plants. The mycoendophytes are currently considered as a new arsenal of secondary metabolites with biological activity. Urtica dioica (nettle common) is considered since ancient times as an important medicinal plant for all its therapeutic properties. To check colonization of nettle leaves by mycoendophytes us colorful trypan blue. We worked on 20 subjects on which we isolated 10 sheets per subject. The sterilization protocol of a leaf surface has eliminated epiphytic fungi. The sheets are cut into fragments which were cultured on PDA medium. Each fungus growing on the fragments is isolated and purified. In parallel, the antibacterial activity was detected against the following strains: E. coli ATCC: 25922, Euterococcus feacalis (ATCC: 49452 et 0409p), Pseudomonas aeruginosa ATCC: 27853, Klebsiella pneumoniae ATCC: 4352, Citrobacter freundii ATCC: 8090, Staphylococcus aureus (ATCC: 43300 et 25923), Bacillus cereus ATCC: 10876. The optical microscope observation of colorful nettle leaves reveal a colonization of the different tissues of the leaf by endophitic fungi. The validity of the sterilization protocol being verified, all isolated fungi are mycoendophytes. We isolated 144 isolates with 18% colonization percentage. Of these some isolates showed significant antibacterial activity against the strains tested. Some endophytic fungi isolated from nettle leaves have antibacterial activity; it is therefore necessary for the extraction of secondary metabolites synthesized by these mycoendophytes and test their biological activity.

Keywords: *Urtica dioica,* endophytic fungi, antibacterial activity, secondary metabolites, trypan blue.

# Microbiological diagnosis of *Tinea unguinum* caused by dermatophytes in the area of Annaba

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Abstract

The dermatophytes are moulds which live with depends on the keratin of the cornea layer on the skin and the superficial body growths on the man and the animals. They pose a rather serious problem of health; they are responsible mainly with surface mycosis; Tinea corporis (skin), Tinea pedis (feet), Tinea capitis (hair), Tinea unguinum (nails) and Tinea crurus (groinion), they can also but to very seldom attend major attacks. *Tinea unguinum* is a dermatosis sitting on the level of the nails caused by dermatophytes and other moulds and yeasts. Our study aims to determine the dermatophytic species responsible for Tinea unguinum in the area of Annaba "East of Algeria" starting from the swabs carried out among patients reached of *Tinea unguinum* on the both sex and various age brackets. In this study the microbiological diagnosis is based on the culture of the swabs carried out on the level of the nails infected on Sabouraud medium added antibiotics and antifungal with incubation at a temperature of 27°c during one week, followed by a macroscopic and microscopic examination of 85 examined cases. According to the morphological characters of the fungal colonies and mycelial filaments, we could identify 13 Trichophyton rubrum with 38,46% followed dermatophytic species: by Trichophyton mentagrophytes with 23,07% and 07,69% for the species of Trichophyton rosaceum, Trichophyton spp., Microsporum longeroni, Microsporum ferrugeneum and Epidermophyton floccosum.

Key words: Dermatophytes, Dermatophytosis, Tinea unguinum, Diagnosis.

### Comparative Assessment of Genotyping Methods for Weissella confusa strains

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### Abstract

*Weissella confusa* is among the most frequently species of the genus *Weissella*. It has been isolated from diverse ecological niches. In this study, a total of 34 *Weissella confusa* isolates from environmental sources (feces, intestinal tract of insects, plants and the rhizosphere) were investigated for molecular intraspecific diversity, respect to the source of isolation. The isolates were analyzed using different genetic fingerprinting molecular methods including, *BOX-PCR* typing and Pulsed field gel electrophoresis (PFGE) analysis. The results of the various methods revealed high intraspecific diversity. BOX-PCR resulted in distinct bands with profiles of great complexity. The profile BOX-PCR revealed six groups of banding patterns, with 27 different genotypes and indicated a significant correlation between the BOX-PCR profiles and the origin of the isolates. *Analysis* of *SmaI-PFGE DNA fragments* showed 28 distinguished pulsotypes and revealed four clustering patterns for environmental isolates. Overall, PFGE was slightly more discriminant method for typing *W. confusa*. This study reported the presence of a specific distribution of *W. confusa* strains according to the source of isolation.

# Specific detection of antibodies by ELISA *Salmonella* Dublin in the milk of cows of the Algiers region

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#### Abstract

*Salmonella* Dublin is a food-borne zoonotic bacteria can be responsible for a serious enteric disease in humans. In pregnant cow, it is most frequently involved in abortions. The transmission of cattle to human can occur in several ways such as the consumption of contaminated meat (especially ground beef) and consumption of milk or pasteurized dairy products or not. The objective of this study is to look for antibodies specific *Salmonella* Dublin in the milk of cows of the Algiers region through the use of ELISA. Analysis of 91 individual samples of cow's milk its showed a prevalence of 13.33%. The collaboration of all veterinary practitioners to reduce the risk of transmission of this pathogen between farms is more than necessary. Indeed, S. Dublin can be difficult to treat and can be fatal in some cases, represents a significant risk to public health.

Keywords: Prevalence, Salmonella Dublin, Cattle, Milk. ELISA

# Effect of quercetin on serine β-lactamase (penicillinase: EC3.5.2.6) from *Bacillus cereus*

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### Abstract

Quercetin is a flavonol belonging to a group of polyphenolic substances known as bioflavonoid. It is typically found in many plants as a glycone or carbohydrate conjugate. Like many other bioflavonoids, Quercetin has many biological properties. Bacillus cereus produces potent β-lactamase conferring marked resistance to β-lactam antibiotics.To overcome this resistance,  $\beta$ -lactam are often given with  $\beta$ -lactamase inhibitors such as clavulanate. In efforts to find new bioactive  $\beta$ -lactamases inhibitors, this study evaluate the inhibitory effect of Quercetin, on serine  $\beta$ -lactamase (penicillinase: EC3.5.2.6) from *Bacillus* cereus in comparaison with clavulanate. Sensitivity of penicillinase activity to the clavulanate and Quercetinwas studied in vitro by using penicillin G as a substrate at 30°C in 1ml of assay buffer (0.1M phosphate; pH7; 0.005mg BSA/ml). The kinetics studies showed that the michaelisconstant (K<sub>M</sub>) and maximum rate (Vm) are 23.78µM and 546.43UI, respectively. These results reflect affinity and high efficiency of penicillinase on penicillin G. IC<sub>50</sub> value that determined by linear computerized regression analysis after logit/log transformation, of clavulanate is 7.24µM; while, of Quercetin is 186.20µM. the comparison of both IC<sub>50</sub> reflect a weak inhibitory effect, which caused by sterically hindered. However, Quercetin is bioactive and exerts its inhibitory effect on penicillinase in a dose-dependent manner; at 140µM, exhibited its most potent inhibitory activity with inhibition percentage of 37.59±0.03%.

**Keywords:** Serine β-lactamase, Quercetin, *Bacillus cereus*, penicillin.

#### Detection of CTXM-15 producing *E. coli* isolates and multidrug-resistant Enterobactericeae in infected poultry in Tunisia

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#### Abstract

The prevalence of extended-spectrum beta-lactamase (ESBL) producing Enterobacteriaceae (ESBL-Eb) isolates has been studied in infected poultry in Tunisia and recovered isolates were characterized for the presence of other resistance genes and integrons. One hundred ten fecal and tracheal samples of poultry (47 tracheal samples and 63 fecal samples) were obtained from 2 farms in Tunisia in 2014. Samples were inoculated onto MacConkey agar plates supplemented with cefotaxime (2 mg/L) and with imipenem (1mg/l) for respectively cefotaxime-resistant (CTX(R)) and (IMP(R)) Enterobacteriaceae recovery. CTX(R) Enterobacteriaceae isolates were detected in 25 out of 110 samples (23%). and one isolate per sample was further characterized. No IMP (R) isolates was detected. The molecular identification of Enterobacteriaceae isolates was performed by sequencing of 16s for the lactose negative isolates and by amplification of *iuaD* gene for the lactose positive. Three species of Enterobacteriaceae (CTX-R) was detected: E. coli (=17), Enterobacter cloacae (n= 4), Proteus mirabilis (n=2) and two Pseudomonas aeruginosa. Only 9 ESBL producing E. coli isolates was identified. The following beta-lactamase genes were detected: bla(CTX-M-1) (six isolates), bla(CTXM-15)+ bla(TEM-1b) (four isolates). The ISEcp1 sequence was found upstream of *bla*<sub>CTX-M</sub> genes in 9 of 10 strains (in three cases truncated by *IS26*), and *orf477* downstream. Class 1 integrons were detected in four strains and contained different genes cassette. Most isolates tested showed a multiresistant phenotype with higher resistance rate for tetracycline (82%) following of sulfamides (70%) and quinolones (64.5%). The clonal diversity will performed by PFGE and MLST method. Chicken farms is not a reservoir of IMP- E. coli isolates however it constitute a reservoir of ESBL-E. coli isolates producing CTX-M-15 class (related of clinical environment) and Enterobacteriaceae isolates (CTX-R) that potentially could be transmitted to humans via the food chain or by direct contact.

#### Isolation and selection of mycotoxigenic fungal strains from apples produced in Eastern Algeria

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#### Abstract

It is known that fruits are commercially and nutritionally important and indispensable food. They play a vital role in the daily human diet by providing the necessary growth factors such as essential vitamins and minerals that contribute in maintaining the health status at appreciable levels. It is estimated that 20-25% of harvested fruit are achieved by pathogens in post-harvest handling, even in developed countries (Droby, 2006; Zhu, 2006). Fungal contamination of fruit can occur during growing, harvesting, transport and storage. Indeed, fruits contain high levels of sugar, nutrients and pH values slightly acid, which makes them a preferred target fungal rot (Singh et Sharma, 2007). Several studies show the isolation of fungi from damaged fruit (Stinson et al., 1981). During refrigeration certain molds persist and may even produce mycotoxins (Tournas et Stack, 2001). Thereby, the objective to which converges this work is to make a mycological study of apples produced in the region of ARIS, city of Batna, especially, isolation and purification of mycoflora contaminating the fruit, followed by mycotoxicological analysis to find and quantify secreted mycotoxins. Therefore, and more frequent contaminants as Aspergillus Niger, and Rhyzopus Muccor, the genus Penicillium strains were obtained. To highlight the power contaminant of the strains obtained, in vivo test on healthy apples has allowed to implement the virulence of three strains of *Penicillium sp.* This virulence is shown by the appearance of damage at fruit tissues inoculated, by selected isolates. . These latter were selected for production of secondary metabolites, which will be the identification object in the next steps of this research, and immuno suppressant effect of the most dominant mycotoxin will be studied in vivo.

Keywords: mycoflora of apples, *Penicillium* sp, mycotoxins, Secondary metabolites.

#### Potential use of chitosan as an antifungal agent

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#### Abstract

For a long time, agriculture across the world was dependent on pesticides. Today, it is influenced by a movement that favours practices that are more sustainable and environmentally safe. To meet these new demands, producers wishing to exploit natural resources and make them profitable must turn to new agronomic practices that combine culture performance and protection at a low environmental cost. In this context, the development of biological molecules able to protect our natural resources is a strategy that is attracting more attention..Natural and healthy for the environment since it comes from the shells of shellfish, chitosan is recognized in several fields and areas of activity, especially for its antifungal properties. Currently used extensively in agriculture, it is possible to think that chitosan can also be used in forestry to meet several needs. This study helps to highlight the antifungal properties of chitosan at different concentrations against Aspergillus Niger. The results show a highly significant inhibition at a concentration of 1 g / 1.

Key words: Chitosan, biomolecule, Aspergillus niger, Environment

## Biochemical and molecular identification of *Campylobacter* strains isolated from fecal sample of poultry in Tunisia

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#### Abstract

The incidence of *Campylobacter* intestinal infections is currently high, and still probably underestimated due to the limited use of detection tests and to problems associated with conditions and cost of the bacteria's culture. However, any medical laboratory is able to diagnose a Campylobacter infection, and the three most frequent species (C. jejuni, C. coli and C. fetus) can be identified using simple criteria. Campylobacter is now recognized worldwide as a leading cause of bacterial gastroenteritis in humans. Campylobacter species are common commensal in intestinal tracts of poultry and livestock, and food products of animal origin are frequently associated with reported cases of illness. The isolation was performed on selective media from 327 fecal samples of poultry, after a pre-enrichment step Bolton Broth for 48 hours at 42°C. Karmali agar was used for the selection of *Campylobacter*. The collection of 110 isolates was characterized and identified by a variety of conventional biochemical tests: Gram stain, microscopic observation search for urease, indole production, production of oxidase, glucose and fermentation of lactose, gas production. Analysis of the results showed that 110 isolates selected present the biochemical and morphological characteristics of Camplylobacter species. The Molecular identification was carried using two primers specific of species respectively for C.jeujeni and C. coli and one universal primer amplifying the *fla* A gene. Only 17 isolates harboring the *fla* A gene specific of Campylobacter and the map gene of C.jeujeni. Amplifincation and sequencing of universal *flaA* gene will be performed for the rest of the collection. Poultry can constitute a reservoir of *Campylobacter* causing infection that potentially could be transmitted to humans via the food chain or by direct contact.

## Isolation of a local strain *Lactobacillus casei* with an inhibitory ability toward *Staphyloccus aureus* and *Pseudomonas aeruginosa* by biofilm formation

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#### Abstract

Antagonistic activity of Lactic acid bacteria toward patogenic germs is assured by secondary metabolites with different inhibitory paths. The aim of this work is to isolate a local strain of lactic acid bacteria with an inhibitory activity by the use of a biofilm without separating the different inhibitory paths, against a clinically isolated strain of Staphylococcus aureus and Pseudomonas aeruginosa ATCC9027, both having the capacity to form a harmful biofilm. The Lactobacillus casei strain was isolated from cow's raw milk and the strain was tested for its ability to form a biofilm. The lactic acid strain was plated with each pathogenic germ on trypticase soy broth in a 96 well microplate. Viable cells were enumerated on specific media using the supernatant at 0h - 14h - 24h - 48h and incubated at 37°C for 24h; staining wells by Cristal violet allowed to bring out biofilm's formation. The inhibitory activity of Lactobacillus casei is stronger against Staphylococcus aureus within 24h (3.3 10<sup>6</sup>ufc/ml for the germ at 0h and 0.25 10<sup>6</sup> ufc/ml after 48h) with an obvious static effect. The pathogenic strain seems to inhibit very weakly the lactic acid bacterium. The Lactobacillus casei appears to inhibit the biofilm's maturation of *Pseudomonas aeruginosa*; however, considering the persistent and adaptive characteristic of Pseudomonas aeruginosa dealing with stress, this inhibition is temporary and not very effective.

**Keywords:** *Lactobacillus casei*, Biofilm, Inhibition, *Pseudomoas aeruginosa*, *Staphylococcus aureus*.

### First detection of NDM-1 producing *Acinetobacter baumannii* in Annaba hospitals – Algeria

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#### Abstract

The antibiotic resistance in Acinetobacter baumannii remains a significant problem in Algeria and can become a major clinical concern in the future. The objectives of the present study were therefore to investigate the prevalence and molecular support of carbapenem resistance in Acinetobacter baumannii. A total of 30 Acinetobacter baumannii were isolated from patients hospitalized between March 2013 and March 2015 in Annaba hospitals . These strains were identified using API 20NE and confirmed using a mass spectrometry (MALDI-TOF MS ). Antimicrobial susceptibility was performed by the disk diffusion as recommended by the susceptibility of the committee of the French Society of Microbiology and E-test Method . Isolates resistant to carbapenems were subjected to different phenotypic test such as modified Hodge test, EDTA test, and modified Carba NP test. Presence of carbapenemase genes was established by PCR and confirmed by sequencing using primers encoding the genes for carbapenemase (bla<sub>NDM-1</sub>, bla <sub>OXA-23</sub>, bla<sub>OXA-58</sub>, bla<sub>OXA-24</sub>, bla<sub>VIM</sub>). All imipenemresistant A. baumannii were positive using the Hodge test and Carba NP test suggesting carbapenemase production and 2 of them were positive with EDTA test .Acquired OXAcarbapenemase-encoding genes were present in all most of A. baumannii isolates, including  $bla_{OXA-23}$  (n = 5) and only two isolates were positive for the  $bla_{NDM-1}$ . Here, we report the first detection of *bla*<sub>NDM-1</sub> in *A. baumannii* in Annaba hospitals - Algeria.

Key words: Acinetobacter baumannii -carbapenemase- NDM-1 -Annaba.

#### Burn Patients Infected with Metallo-Beta-Lactamase-Producing *Pseudomonas aeruginosa*: Multidrug-Resistant Strains

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#### Abstract

Metallo-beta-lactamase producing *Pseudomonas aeruginosa* is a leading cause of mortality & remains a serious health concern among the clinicians. The aim of this study was to detect MBL-producing *P. aeruginosa* in burn patients and determine multidrug-resistant (MDR) strains.

The study was carried out in Burn unit from the period of April 2014 to January 2015. Total of 30 Multidrug resistant (MDR) *P. aeruginosa* isolated were identified by standard microbiological techniques & were tested for Antibiotic susceptibility. Minimum inhibitory concentrations & MBLs were determined by the E-test method. Screening of carbapenemase producer isolates was performed by using imipenem-EDTA double-disk synergy test & modified Hodge test. The presence of genes encoding carbapenemases was investigated by PCR. The clonal relatedness among isolates was analyzed by PFGE method. In antibiotic susceptibility testing by E-test method 17 isolates (55%) were resistant to all the 11 tested antibiotics. An imipenem-EDTA synergy test was positive for 7 out of 17 (22.58%) strains resistant to imipenem that were PCR positive for VIM-4 carbapenemase. The characterization of class I integrons showed a class I integron that harbored the VIM-4 gene associated with gene encoding aminoglycosides resistance *aadA7*. Careful of antibiotics prior to prescription may help to prevent, treat, & control MDR *P. aeruginosa* strains in burn units.

Keywords: Burn; P.aeruginosa; Metallo-Beta-Lactamase; Drug Resistance

# Serogrouping, antibiotic resistance and detection betalactamase extended spectrum (ESBLs) strains avian pathogenic *Escherichia coli* responsible for colibacillosis in broilers in East Algeria

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#### Abstract

The aim of this work was to search among the strains of *E.coli* isolated from broiler chickens suffering of colibacillosis those producing ESBL, and determine their serogroups and to assess the frequency of resistance of these strains to 11 molecules antibiotics. One hundred E. coli strains were isolated from the livers and spleens of animals on MacConkey agar, and biochemically identified by API 20 E system (Biomerieux, France). The susceptibility testing is performed according to the disk diffusion method on Muller Hinton agar. Search ESBL strains was done by applying the AMC disk at 30 mm of a disk of C3G (cefotaxime ) and confirmation by the technique of double disc, serotyping was done by rapid agglutination blade technique (ARL) sera used are O1, O2, O78 (Biovac, France), which are known pathogens. Our results show very high percentages of strength vis-à-vis amoxicillin / Ac Clavulanic (92%), ampicillin (89%), nalidixic acid (99%), sulfa-sulfamethoxazole (82%), enrofloxacin (82%), and tetracycline with 100% of resistant strains. All strains are multirésitantes, all resistant to at least two antibiotics. We also isolated 03 strains that are producing ESBL. The dominant serotype O2 55% of strains are followed by 16% and O78 O1 avec14% while 15% of non-typeable strains (NT) by these sera. In conclusion, it is clear that antibiotics are becoming less effective against E. coli, in addition to the emergence of APEC strains that are producing ESBL is needed more than ever to think of an alternative to antibiotics.

Key words: Algeria, antibioresistance, colibacillosis, Escherichia coli, serogrouping

#### The decontamination of water by the removal of antibiotics with a membrane process

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#### Abstract

The study aims to decrease water pollution. We directed this work for the recovery of the antibiotics (penicillin V) in the industrial wastes. Technologies of the membrane processes are very known in last years by their effectiveness of recovered, concentrated the solutions in suspension. The assembly of a frontal microfiltration as puted in work for this study with an acetate membrane of 0.2 µm and for constant pH of the initial solution. The trace of the t/V report according to the volume of perméat allows calculates it resistances of the cake and support for different value from concentrations and transmembrane pressure. The experimental results express that the resistance of the cake (deposit) becomes more important when the transmembrane pressure increases, and that the flow of the permeat decreases considerably with time and increases with the transmembrane pressure.

**Key words**: Frontal microfiltration, penicillin V, membrane acetate, filling.

#### Evaluation of the effectiveness of oxytetracycline against the bacterium *Paenibacillus larvae* a causative agent of American foulbrood

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#### Abstract

American foulbrood is a more serious bacterial diseases bee Apis mellifera. The causative agent Paenibacillus larvae, has a strong spread of power Oxytetracycline (OTC) is the most widely used active ingredient for the treatment of this disease in Algeria. The objective of this study was to evaluate the prevalence of the bacterium *Peanibacillus larvae* causative agent of American foulbrood in apiaries of some regions in central Algeria and determine the effectiveness of oxytetracycline against this bacterium in laboratory. Microbiological, biochemical and microscopic tests were used to assess the prevalence of this pathogen. The majority of study areas infested by this pathology with an average rate of 47.56% of contamination. We have established the 15 isolates resistance profile vis-a-vis oxytetracycline by the disc diffusion method. The classification of inhibition diameters in decreasing order showed that 8 of 15 samples are resistances. The majority of resistant strains are located in two regions of Blida and Tizi Ouzou. To avoid the appearance of this phenomenon of antibiotic resistance poses a major problem for the health of bees, it is best to alternate the use of different families of antibiotics. It is also about putting a program of prevention and control of this disease by veterinary authorities to reduce the risk of the spread of this disease to other apiaries.

Keywords: Paenibacillus larvae, resistance, prevalence, Algeria.

## Isolation of bioactive actinomycetes from soil samples collected in water source in the region of north Algeria

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#### Abstract

The constant evolution of bacterial resistance to antibiotics and the emergence of new infectious diseases is a major public health problem; hence the urgent need for new antimicrobial molecules. Ten soil samples were collected in and around a source known as "Amezyav ath yaou" located in a forest area in the region of Tizi-Ouzou (northern Algeria) within the framework of the search for novel bioactive compounds producing actinomycetes strains. Three isolation media were used (Bennett, GLM and Sabouraud). A total of 27 strains of actinomycetes with different morphological features were isolated. A significant difference in the number of colonies was observed between the different culture media. After purification, the antibacterial activity of the twenty-seven actinomycetes strains was tested against three bacterial strains from the ATCC collection (Escherichia coli ATCC 25922, Pseudomonas aeruginosa ATCC 27853, and Staphylococcus aureus ATCC 25923) by two agar diffusion methods: the perpendicular streak method and the agar cylinder method. Among 27 isolated strains, 13 showed antibacterial activity toward at least one bacterium in the primary screening. Of the 13 strains showing an antibacterial activity, 5 of them were found to be highly active against *Staphylococcus aureus* with inhibition diameters ranging from 20 to 26 mm. The five isolates were then selected for identification. The results are very encouraging and may suggest that actinomycetes possess molecules that predict their application against multi-resistant bacteria.

**Key words:** Actinomycetes; Antibacterial activity; Isolation; Bioactive compounds; Multiresistant bacteria.

#### Pentachlorophenol effect in biofilm formation and antibiotic sensibility

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#### Abstract

Through chemical contamination of natural environments, microbial communities are exposed to many different types of chemical stressors; however, research on responses to this contaminant stress is limited. The existence of these chemicals in groundwater, industrial wastewater effluents, sediments, and surface soils poses great challenges for treatment and remediation. Pentachlorophenol (PCP) is one of many recalcitrant and toxic compounds found in water that are used for various purposes, such as herbicides, insecticides, fungicides, wood preservatives, resins, and lubricants. This study examined PCP effect in the biofilm and the antimicrobial sensibility response by consortium bacterial. To elucidate the effect of PCP on the microbial activities of biofilms, we used two methods to evaluate the activation caused by PCP. Biofilm producing strain was determined by various methods to found the PCP effect for two bacterial consortiums. In the current study, Congo Red Agar methods and Microtitre plate biofilm formation assay were used (in different PCP concentration). Antimicrobial resistance of this isolate was determined by the agar disk diffusion method. Antimicrobial resistance of these 8 isolates (2 consortiums) was determined by the agar disk diffusion method. The bacterial biofilm response is more important in a bacterial consortium than in a single strain also PCP contaminant decrease antimicrobial sensibility.

Keywords: Biodegradation, Pentachlorophenol, biofilm, Antimicrobial sensibility, consortium

## Phenotypic and molecular identification of antibiotic resistance genes of bacterial strains isolated from mastitic milk in Algeria

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#### Abstract

The objective of this study was out to investigate the phenotypic and genotypic identification of *in vitro* antimicrobial susceptibility of 21 Staphylococci (10 *Staphylococcus aureus* and 11 Coagulase Negative Staphylococci) isolated from bovine mastitis to 12 antimicrobial drugs frequently using in veterinary medicine in Algeria. Isolates were tested for antibiotics with disc-diffusion method according to the National Committee for Clinical Laboratory Standards guidelines in the Mueller-Hinton agar, and resistant genes *mecA*, *blaZ*, *aac-aph*, *ermA*, *ermC*, *tetK* and *tetM* were detected by PCR. Staphylococci isolates showed high resistance to penicillin (95.23%), oxacillin (80.95%), clindamycine (80.95%), and erythromycin (76.19%) but, no resistance of all these strains was detected for gentamicin. Among 21 isolates of Staphylococci, 20 were found to be methicillin and multidrug resistant. The distribution of antibiotic-resistant genes was *mecA* (100%), *tetM* (100) followed by blaZ (42.85%). In the present work, the significant determination was the high prevalence of methicillin-resistant Staphylococci. The finding of methicillin-resistant staphylococci (MRS) from bovine mastitis is the first report in Algeria and revealed the status of resistant isolates in herd that might be helpful in treatment, controlling of resistant strains and for deciding culling of cows.

Key words: Antimicrobial susceptibility, Bovine mastitis, Resistance genes, Staphylococci.

#### Escherichia coli resistant to β-lactams isolated from feces of Broilers chickens

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#### Abstract

In our study we focus on farms around a radius of 40 km from the city of Tlemcen. From April to June 2014, we collected 140 feces samples from housing of healthy broiler chicken flocks at 4 different rearing sites in Wilaya of Tlemcen during fattening in the rearing period for each farm. Each of the 4 flocks comprised between 800 and 40.000 birds per house. The specimens were collected by walking through the housing. The result was 34 non-duplicate strains of E. coli were isolated from 2 farms. Antimicrobial susceptibility tests of these strains were established by the agar diffusion method according to the standard operating procedures the Comité de l'Antibiogramme de la Société Française de Microbiologie (CA-SFM) 2013 guidelines, using Mueller-Hinton and adjusted inoculum to 0.5 on the scale McFarland and further diluted to 1/100, the panel consisted of amoxicillin, ampicillin, ticarcillin, amoxicillin/clavulanic acide, ticarcillin/clavulanic acid, piperacillin, aztreonam, ertapenem, cephalothin, cefuroxime, cefamandole, cefotaxime, ceftazidime, ceftriaxone, Moxalactam, cefixime, cefoxitin, tobramycin, amikacin, nalidixic acid, ciprofloxacin, norfloxacin, colistin, and trimethoprim/sulfamethoxazole. These strains show resistance to  $\beta$ -lactams and quinolones. The minimum inhibitory concentrations (MICs) were determined by agar dilution method in Mueller-Hinton medium (Fluka BioChemika, Spain), in accordance with the CA-SFM 2013 guidelines. The method of serial dilution in a solid medium is used to determine the bacterial susceptibility to antibiotics. It consists in a standardized bacterial inoculums contact with increasing concentrations of antibiotics. E. coli strain ATCC 25922 was used as a control.

Keywords: E. coli, Antibiotic resistance, feces, broiler chickens.

#### Comparison of three Asteraceae species from Algeria for their in vitro antimethanogenic effect

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#### Abstract

Tree plants (Chamaemelum nobile, Chrysanthemum sengetum and Centaurea Pullata) were tested to determine *in vitro* their effects as feed additives to decrease ruminal methanogenesis. A control diet (50% of alfalfa hay, 20% of ryegrass hay and 30% of corn) additionned with each plant or without addition were incubated (39°C for 24h anaerobically) with buffered rumen fluid. At the end of incubation, pH, gas and methane production, DM digestibility, Total volatiles Fatty acids (TVFA) and methane-forming Archaea were measured. All the plants tested reduced methane production (P < 0.01) in relation to the control (no plant added). C. sengetum and C. pullata caused the highest reduction in methane production (36.5% and 28%, respectively), whereas C. nobile shows less effect on methane reduction (15.2%). Gas production, pH and fermentation efficiency (PF) were not affected by any plants addition (P > 0.05). Among the plant species evaluated, Chrysanthemum sengetum and Centaurea Pullata showed the most consistent effects for decreasing methane production without adversely affecting other parameters of rumen fermentation, since it showed the greatest and most consistent effect on methane production (-36.5% and -28%) in relation to the control, (P < 0.001). Moreover, it decreased DM digestibility (P < 0.01) and methanogen Archaea in the rumen.

## Effect of alkaloids of *Nigella sativa* and *Nigella arvensis* against soft rot induced in two varieties of potato (*Solanum tuberosum*)

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#### Abstract

The potato crop (Solanum tuberosum) and the harvested tubers are subject to many bacterial aggressions such as *Clavibacter*, *Pectobacterium*, *Ralstonia*, and *Streptomyces*. Latter include bacteria belonging to the genera *Pectobacterium* and *Dickeya* (Dk), more known as *Erwinia carotovora*. These bacteria are responsible for extensive damage during cultivation and postharvest conservation of potato tubers. Today, no treatment can effectively fight against this disease. The objective of this work is to evaluate, in vivo and in vitro, the effect of alkaloids isolated from *Nigella sativa* and *Nigella arvensis* in protection against soft rot in two varieties of potatoes (Spunta and Desiree) generated by three strains of pectinolytic bacteria (Pectobacterium carotovorum subsp. atrosepticum (Pca), Pectobacterium carotovorum subsp carotovorum (Pcc) and Dk). The results show, in general, the alkaloids isolated from the seeds of *Nigella sativa* and *Nigella arvensis* significantly reduce soft rot rates in two varieties Desiree and Spunta with maximum reduction of about 83%. When the test is carried out on the variety Désiree, alkaloids of Nigella arvensis appear more effective against Pcc as those extracted from Nigella sativa (78% against 41%), while Nigella sativa alkaloids are more effective on the DK. The situation changes when considering the effects of these compounds on the variety Spunta: Nigella arvensis alkaloids are more effective against DK than Nigella sativa. Results already allow us to say that the variety of potato may interfere with type of molecules.

Key words: alcaloïdes, Nigella sativa, Nigella arvensis, soft rot, Pcc, Pca, DK.

#### Analysis of microbiological properties of raw cow milk in the center area of Algeria

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#### Abstract

Raw or processed milk is known to support the development and multiplication of several microorganisms. This study was conducted in order to assess the microbial load of raw milk produced in Algeria by a pathogenic bacteria considered as toxic. In total 30 samples were collected aseptically from the point of delivery of the dairy state of Boumerdes and analyzed for 6 weeks on 5 tanks of different collectors. Five relevant to the dairy industry namely pathogenic bacteria : Salmonella spp, Listeria monocytogenes, Clostridium sulfit- reducing, Escherichia coli and Staphylococcus aureus were studied. The results showed that 13% of milk were contaminated with *Listeria*. 7 samples were positive for *S.aureus*, germ recognized as a major cause of clinical and subclinical mastitis in dairy cattle farms, with an average count per milliliter of  $1.85 \pm 0.68$  Log10 cfu. The average value of *E.coli* bacteria was  $5.64 \pm$ 1.27 Log10 cfu / ml. The sulphit-reducing clostridia were detected in 9 samples achieved varying levels between 0 and 60 cfu / ml. All samples were free of Salmonella spp. A very large variability was observed in the number and the type of microorganisms detected. It was concluded that the main common risk factor associated with contamination of raw milk by these pathogens is linked to the number of farms (p < 0.05) where milk is collected and to the hygienic level of each farm (p < 0.001).

Keywords: pathogenic germs, tanks, delivery, raw milk.

#### Mechanisms of Resistance to Carbapenems of Gram-Negative Bacilli Isolated from Aures Region: Molecular and Epidemiological approach

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#### Abstract

During these last years, the Gram-negative bacilli have played an increasing role in nosocomial infections with a high epidemic risk having consequences like therapeutic difficulties. Its epidemiology in the world is marked by the release of a majoritaries clones harboring genes encoding resistance to carbapenems. The aim of this study is to evaluate the resistance levels to antibiotics of Gram-negative bacilli, stains isolated from hospitalized patients of «Benflis Touhami» CHU-Batna-, over a period of 02 years (2013-2015). A total, 284 Gram-negative-bacilli were isolated.172 (60,6%) isolates were found to produce ESBLs, 88 strains (31%) showed a complete resistance to carbapenems distributed as follows: (22 Klebsiella pneumonia, 40 Acinetobacter spp, 24 Pseudomonas spp, 1 Esherichia coli and 1 Proteus mirabilis), 8 strains (2,8%) hyper-product their Cephalosporinase (CHN), 4 strains (1,40%) present associate mechanisms (CHN+ESBLs) and 2 strains (0,70%) hyper producing AmpC beta-lactamase. It should therefore be emphasized that the emergence of such multidrug resistance strains which is the origin of the epidemic is worrisome because it has limited treatment options for this type of infections. Finally, a strict control policy should be implemented in our hospitals to limit the emergence of multiresistants/totoresistants to antibiotics.

**Keywords:** Nosocomial infections, Resistance to carbapenems, Gram-negative-bacilli, ESBLs, Carbapenemases, Multiresistants/totoresistants.

## Effect of the resistance to cefotaxim on the metallo-resistance of *Escherichia coli* isolated from surface water

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#### Abstract

Wastewater, industrial and hospital discharges play an essential role in the dissemination of antibiotics and heavy metals resistance genes. Our study focuses on the effect of the tolerance to heavy metals on the growth of *E. coli* resistant to cefotaxime (CTX) isolated from surface water of the Oued Bousselam. A serial of two fold dilution was carried out from 51200  $\mu$ g/ml of Cd(NO<sub>3</sub>)<sub>2</sub>, HgCl<sub>2</sub> et ZnSO<sub>4</sub> inoculated with 10<sup>6</sup> UFC/ml of *E.coli* in Mueller-Hinton Broth by microplate. The concentrations for optimal growth for most strains were 160, 5, 320  $\mu$ g/ml, and the MICs were between 640-1280, 5-40 and 1280-2560  $\mu$ g/ml for Cd, Hg and Zn, respectively. Plasmid extraction was realized by Sambrook protocol. However, 24 strains out of 30 have at least one plasmid but two of them containing two and three plasmids separately. The cross-resistance between CTX and heavy metals exist phenotypically, despite the absence of correlation between plasmid profile and multi-resistance in some strains.

Keywords: wastewater, heavy metal, E.coli, cross-resistance, plasmid profils.

#### Isolation and selection of Actinomycetes producing bioactive substances

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#### Abstract

To isolate, characterize and evaluate the potential of *Actinomycetes* in the region of eastern Algeria to produce bioactive molecules. A total of 16 strains were isolated and purified from 4 different semi-arid rhizosphere soils. The antagonistic activity of actinomycetales isolates were tested against Gram positive and Gram negative bacteria as well as the phytopathogenic fungi by agar cylinders technic. The extraction of bioactive molecules from the culture filtrate of the selected strains was performed with ethyl acetate solution and the crude extract was tested by diffusion disc method. A thin layer chromatography was performed on crude extracts. From the 16 actinomycetales strains tested, 13 strains were active against at least one of bacteria test and 15 are active against phytopathogenic fungal strains. CVF strains and LAC1 have demonstrated a significant antagonist activity. The results obtained by the method of agar cylinder are better than those obtained by disc diffusion. Both crude extracts have shown a good activity against phytopathogenic fungi and Gram-positive bacteria. The thin layer chromatography of silica gel technic of crudes extracts CVF and LAC1 revealed the presence of at least two molecules, which has antimicrobial activity. The results of this study showed that Actinomycetes of the semi-arid region of eastern Algeria have an important role to produce molecules with potential antimicrobial activities.

Keywords: *Actinomycetes*, Cylinder agar method, diffusion method by paper disc, extract antibiotic, antimicrobial activity.

#### Mechanism of action of *Cypripedium montanum* and *Daucus gracilis* essential oil on *Bacillus cereus, Klebsiella pneumonia* and *Proteus mirabilis* determined by time-kill assay and cell lysis

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#### Abstract

To avoid the spread of pathogens, synthetic substances have been widely used to diminish microbial growth or inhibit microorganisms. However, because of their side effects, much attention has been paid to use natural antibacterial products. Recently, essential oils have received attention in their useful physiological functions and antimicrobial activity. Two local Apiaceae species have been used for their essential oils obtained by hydrodistillation. Their MICs against three ATCC strains were determined at earlier by the dilution method. In order to determine the lysis mechanism of these oils, two methods were used; the time-kill assay and the cell lysis assay. In the time kill assay, three ATCC bacteria; *B. cereus*10876, *K. pneumonia* 700603 and *P. mirabilis* 35659were exposed to MICs of *C. montanum* and *D. gracilis* essential oil to evaluate the effect on the growth rate of surviving bacteria through time. The bacterial cell lysis was determined by the measurement of the optical density at 620 nm at different time intervals after having incubated the same strains with concentrations of MICs and four times MICs. Time-kill assay showed that essential oils caused a remarkable decrease in the survival rate as well as in the absorbance was recorded, which suggest that the cytoplasm membrane is one of the targets of the essential oil.

## Session 03

## **Adaptations / Biotic Interactions**

#### Keynote 4

# The *Frankia-Casuarinaceae* nitrogen-fixing symbiosis: study of early molecular mechanisms promoting the symbiotic interaction and the adaptation to poor and degraded soils

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#### Abstract

Frankia actinobacteria are soil filamentous Gram+ microorganisms belonging to Actinomycetal order and Frankiaceae family. These bacteria are not an obligate endosymbiont and can occupy two distinct ecological niches, the root nodule and the soil. They are able to fix nitrogen either in a free-living state or in association with actinorhizal plants and can display 3 cell types: hyphae the vegetative structures, spores which are thought to contribute to the dissemination and survival of the microorganism and vesicles specialized in N<sub>2</sub>-fixation. Frankia bacteria are able to develop root nodule  $N_2$ -fixing symbiosis with more than 240 species of non-legume dicotyledonous plants, mostly trees and shrubs, belonging to eight different families and 24 genera. Thanks to their symbiotic properties, actinorhizal plants contribute to improve soil fertility in disturbed sites and grow well under a range of environmental stresses such as high salinity, heavy metal pollution and extreme pH. This facility for adaptation is responsible for the great interest in actinorhizal plants, particularly in several species of *Casuarinaceae* such as *Casuarina glauca* and *C*. *equisetifolia* which are widely planted in the tropics and subtropics to stabilize sand dunes. produce fuel wood, rehabilitate area with industrial waste and to increase soil fertility in agroforestry systems. The basic knowledge of the symbiotic association between Frankia and actinorhizal plants is still poorly understood and the early molecular regulatory events involved in actinorhizal nodulation is limited by several factors such as the lack of genetic approaches for both Frankia and the host actinorhizal plants that are trees and shrubs.Our research is focalized on the study of the Frankia-Casuarinaceae symbiosis and the general objective of our group is to understand molecular mechanisms that allow the development of actinorhizal nodules. This program is conducted in the contexts of the adaptation of the root system to poor and degraded soils and the comparative analysis of actinorhizal symbiosis and Rhizobium-Legume symbiosis. Our works aim to decipher how Frankia signal molecules are perceived and interpreted by the host-plant, thus contributing to promote the symbiotic interaction and the adaptation to poor soils. The ultimate purpose is to select the most efficient and better-adapted Frankia-Casuarinaceae associations. The presentation will provide an overview of our work on the molecular study of Frankia-Casuarinaceae symbiosis and the recent advances in the field will be discussed.

#### Keynote 5

#### Root nodules in *Casuarina glauca* are also induced by non-*Frankia* actinobacteria of the genus Nocardia

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#### Abstract

Frankia are the common actinobacterial symbionts that form nitrogen-fixing root nodules with the actinorhizal plants. Recently several non-Frankia actinobacteria have been isolated from the nitrogen-fixing root nodules of these plants. Two strains BMG11209 and BMG51109 affiliated to the genus Nocardia were revealed to axenically induce root nodulelike structures that highly promote the growth of seedlings of their original host plants Casuarina glauca. We also sequenced and obtained two draft genomes: the 8.78 Mb genome of Nocardia BMG51109 and the 9.14 Mb genome of Nocardia BMG11209. Analysis of both genomes revealed potential biosynthesis of plant auxins. Using Electron Microscopy, the cortical cells of the plants showed to be infested by nocardiae rod-shaped cells. The root hair deformation in transgenic Casuarina seedlings infected by BMG111209 strain was visible over a large zone of the roots with an early ProNin::Gus expression in the infected root cells. Furthermore, co-infection experiments using Nocardia and Frankia have found to induce an early onset of plant nodulation timeline compared to plants infected with Frankia alone. Coinfection also caused an increase in the number of nodules present on the plants. These Nocardia strains together with their nodule-like structures induced in the roots of Actinorhizal Casuarina glauca plants evoke many questions on the ecological significance and functionality of this new discovered symbiosis.

#### Keynote 6

# The impact of Arbuscular mycorrhizal fungi and *Frankia* on *C. equisetifolia* growth and soil microbial catabolism diversity depends on the origin of the symbiotic microorganisms

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#### Abstract

In recent decades, fast-growing trees like Australian trees were introduced in West Africa to recover degraded lands. In order to improve their performances, trees are often inoculated with mycorrhizal fungi but generally poor attention has been given to strains selection, despite the fact that huge variations in the effect on plant growth promotion can be observed between different strains. Strikingly, studies comparing the effect of local strains versus strains from the origin zone of the plant host are very rare. Although these could be potentially very interesting because of the long coevolution between the plant and its fungal partner may have taken place in the zone of origin resulting in an optimization of the symbiosis effectiveness. In this work we focused on the impact of microbial complexes (Abuscular mycorrhiza and Frankia) associated with C. equisetifolia in its origin zone (Australia) and in its introduction zone (Senegal): (1) by studying the impact of a Senegalese or an Australian inoculum on the growth of C. equisetifolia and the response of soil microbial catabolic diversity to arbuscular mycorrizal fungi. Our results show that regardless of its origin, the inoculum has a growth promoting effect, and provokes significant changes in the structural and functional diversity of soil microbial community. This growth promoting effect is increased in plants inoculated with Senegalese inoculum. The origin of the inoculum has an impact on the composition of microbial community, and more diversity is found among Australian samples.

Key words: Casuarina equisetifolia, Arbuscular mycorrhizal fungi, Frankia, Diversity, Bacterial community

## **ORAL COMMUNICATIONS**

#### Effects of two microbial organisms (fungus and bacterium) on the desert locust Schistocerca gregaria

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#### Abstract

We studied the efficacy of two biological products from fungi and bacteria origins to treat the desert locust *Schistocerca gregaria* imagos. The entomopathogenic fungus *Metarhizium acridum* showed a great toxicity with both tegumentary and oral way. In the contact treatment, the efficacy begins in the first days and the mortality reaches 100% in 7 days. On the contrary, in the ingestion treatment, the mortality just begins in the 7<sup>th</sup> day and reaches 100% in the 9<sup>th</sup> day. The histological study on the locust's gut treated by ingestion showed lesions at all levels. Therefore, the fungus *M. acridum* can grow in both the locust's gut and tegument. The other biological product that we tested; the bacterium *Bacillus subtilis* subsp. *Spizizenii* ATCC 6633 was not effective. The histological study on the locust's gut treated by this bacterium did not show any tissue alteration.

Keywords: Schistocerca gregaria, Metarhizium acridum, Bacillus subtilis, Microbial organisms, Entomopathogenic fungus.

## Effects of plant growth promoting rhizobacteria associated to citrus tree to alleviate plant drought stress

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#### Abstract

Water deficiency is one of the most serious environmental abiotic stresses. Prolonged drought stress induces severe constraints for plant health and productivity. Plant growth promoting rhizobacteria (PGPR) have been shown to play an essential role in improving the plant growth and the crop yields under stressful conditions. In this context, this study was aimed to evaluate the effects of citrus rhizobacteria, exhibiting high level of PGP features, on plant growth under drought stress. Six drought tolerant PGPR namely *P. japonica, P. jessenii, P. resinovorans, P. poae* and two *Ensifer adhaerens* strains were inoculated on tomato and pepper plants subjected to drought stress during 14 days. The growth parameters; shoot and root lengths, shoot and root fresh and dried weights and number of leaves were measured., Among the tested rizobacteria, one strain of *Ensifer adhaerens* has shown the highest effects on growth parameters for the two investigated plants. This study highlighted the efficient use of *Ensifer adhaerens* as biofertilizer, under drought stress condition, for the enhancement of plant growth.

## Image analysis and quantitative PCR method for rapid assessment of fungicide resistance in plant pathogens

#### Semcheddine Cherrad, C. Hernandez and S. Vacher

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#### Abstract

For several years, in agricultural practices and crops overproduction strategies fungicides have become a major component of plant disease control and management. Long-time application of same fungicides in field led to fungicide resistance emergence. With fewer fungicides available (particularly with new modes of action) and increasing consumption, risk of fungicide resistance is even greater, and failures will more profoundly impact cropping systems (Beckerman, 2013). All these factors increase the need for rapid detection and assessment of fungicide resistance to help growers to manage disease control. Laboratory tests are performed on field isolates to investigate potential and level of resistance. In most case, choice of test depends on several parameters like biology of plant pathogen and mode of action. In addition to classically biological tests, we developed new image analysis test offering rapid and accurate measure of mycelium or germinate growth of fungi in selected medium supplemented with fungicides molecules. Software image analysis of elongated germinate tube allow us to evaluate ratio of resistant's spores in field isolated population. Many fungi, including the economically important powdery mildews are obligate biotroph. Genetic testing of potentially resistant isolates was performed directly from disease lesions, obviating the need for isolation, and subsequent growth of the fungus in vitro. Developed quantitative real-time PCR (qPCR) has been applied to the detection and quantification of microorganisms resistant to QoI (Quinone outside inhibitor) and DMI (demethylase inhibitors) fungicides. These data help to rapidly monitor resistant development in crops and manage disease fungicide control programs.

#### Preview of bacterial community in the actinorhizal tree *Casuarina* sp. Phytosphere

#### Amir Ktari, Karima Hezbri, Faten Ghodhbane-Gtari, Wajdi Mimouni, Abdellatif Gueddou, Moussa Louati, Imen Nouioui, Abdellatif Boudabous, Maher Gtari\*

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#### Abstract

*Casuarina* sp. are woody trees best known as fast growing and highly tolerant to drought and salinity under tropic and subtropic climates thanks to their ability to establish several symbiotic relationships with mycorrhizal fungi and with the nitrogen-fixing actinobacteria Frankia. The bacterial community of Casuarina sp. phytosphere was examined using both culture-dependent and -independent methods targeting endophyte and epiphyte bacterial communities. Samples (leaves, stems, roots and nodules) were collected from mature plants growing in Tunisia. Denaturing Gradient Gel Electrophoresis (DGGE), ITS 16S-23S rRNA PCR-profiling and partial 16S rRNA genes sequencing from isolated strains were used to identify the bacterial community present. On the basis of combined data set of 385 16s rRNA gene sequences obtained from 126 isolated strains dereplicated from a collection of 330 isolates together with 55 sequenced bands from PCR-DGGE, 8 phyla classified into 45 genera and 117 OTU (Operational Taxonomic Unit) were recorded across Casuarina phytosphere. Statistical analysis showed that the lowest diversity was controversially harbored by nodules, while roots are the most rich followed by stems and leaves. Based on phylogenetic analyses, Actinobacteria is the most dominant phylum (21.04-100%), followed by Proteobactertia (11.11-65%), Firmicutes (4-63.5%), Bacteroidetes (1.3-12%) and Deinococcus-Thermus (0.7-2%) phyla. Endophytic bacteria were retrieved from all plant tissues with dominance of actinobacterial genera such as *Streptomyces*, Micromonospora, Nocardia and Frankia in root nodules. The noteworthy bacterial richness and diversity revealed sides of the successful adaptability of one of the most important and widely introduced actinorhizal species under tropical and subtropical climates.

#### ABI-05

## Physico-chemical and microbiological characterization of steppe soils (region of El Bayadh in Algeria)

#### Fatima Zohra Abdi<sup>1</sup>, Nadia Benouaz<sup>1</sup>, Wahiba Moumen<sup>1</sup>, Khalladi Mederbal<sup>2</sup>

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#### Abstract

The steppe region is oriented mainly pastoral, with a strong pressure anthropozoic resulting in reduced biological potential. To contribute to the understanding of how these ecosystems work was under taken at the interface region steppe-Saharan region (the wilaya of El Bayadh). This work has allowed the characterization microbiological and physico-chemical soil. The study focused on six (06) stations distributed in a north-south transect in the axis-El Bayadh-Brezina. The dominant plant species are *Erucavescicaria*, *Limonium scoparium Sinuatum* and *Arthrophytum*; the *Atriplexe canesces*, *Hordeum*, *Enarthrocarpus clavatus* and an artificial forest of *Aleppo pine* in the stations (St1, St2, St3, St4, St5et St6, respectively). Preliminary microbiological analyzes show the presence of bacteria very different from one station to another, these results showed the dominance of groups of bacteria *Pseudomonas*, *Staphylococcus*, Sulfitoreducers, Enterobacteriaceae and *Streptococcus*. In addition, the results of physico-chemical analyzes of soil when sampling was conducted can see the predominance of calcareous alkaline pH, with a moderate level of salinity. We note that the results are consistent and can establish a relationship between bacterial groups and the content of organic matter, water holding capacity and land.

Keywords: steppe region, soil, microbial and physico-chemical analysis, El Bayadh.

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## Characterization of microorganisms associated with Algerian honeybee: *Apis mellifera intermissa* and *Apis mellifera sahariensis*.

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#### Abstract

The microorganisms associated to the two Algerian subspecies honeybees *A.m.intermissa* and *A.m.sahariensis* were characterized. The identification of the microflora associated with the digestive tract and pollen was made two seasons during the year 2012, namely in January - February (winter test) and May (spring test). The research of this microflora was realized by the digestive tract, stored pollen (bee bread) and pollen baskets. The Isolated bacteria belong to the genera *Enterobacter, Pantoea, Pseudomonas,* and *Lactococcus* and to genera neighbors, *Lactobacillus* and *Streptococcus*. All yeast strains are likely to belong to the Saccharomyces genera and the molds are identified genera *Penicillium, Aspergillus, Cladosporium, Alternaria* and *Mucor*. According to the biochemical and physiological characters that are found, certain bacteria may be involved in the degradation of the components of the two extern layers of pollen, be ingested by the bees with food and shared with other individuals of the hive by the phenomenon of trophallaxis. The presence of yeasts and molds can be explained by the close relationship as maintenance honeybee with its environment, soil and flowers.

Key words: *A.m.intermissa, A.m.sahariensis*, microflora, digestive tract, bee bread, pollen baskets, bacteria, yeasts and molds.

## POSTERS

#### ABI-P1

#### Antimicrobial activity of endophytic actinomycetes from roots of *Aretemisia absinthium* in Algeria

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#### Abstract

The endophytic are bacteria responsible for the production of most bioactive molecules. Research works related to the isolation of these microorganisms from medicinal plants of Algeria are rare. The objective of this work is the isolation of these endophytic actinomycetes. Nine strains of endophytic actinomycetes were isolated from the roots tissues of *Aretemisia absinthium* of Batna (Algeria ). Antimicrobial activity was tested against four Gram-positive bacteria, four Gram-negative bacteria and one yeast using the agar cylinder method. All strains showed antimicrobial activity against at least three of the test-germs studied.

Keywords: Endophytic actinomyctes, *Aretemisia absinthium*, antimicrobial activity, antimicrobial molecules.

#### ABI-P2

## Sublethal effect of bacterium *Bacillus thuringiensis var kurstaki* on the development of *Culex pipiens* (Diptera, Culicidae)

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#### Abstract

The bacterium *Bacillus thuringiensis var krustaki* (*Btk*), is one of the most used insecticides at present as a biocontrol agent to suppress populations of various insects harmful. This framework, we examined power on larvae the 4th stage of the mosquito *Culex pipiens* (Linee) in laboratory conditions. Larvae were exposed to different concentrations of Btk (25mg / 1 50mg / 1 83mg / 1). The larvae of *Culex pipiens* were highly susceptible to *Btk*; and chemical analysis by gas chromatography (GC) shows that the product is on the quantities of culcular waxes in emerging adults of this treatment.

Keywords: Bacillus thuringiensis var krustaki, biological insecticide, Culex pipiens

#### ABI-P3

### Contribution to study the biodiversity of rhizobia nodulating *Retama raetam* and *Lupinus* sp. in Algeria

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#### Abstract

Legumes are one of the largest groups among the plants. These play a key role in the conservation of plant cover. Their root systems, especially, making them ideal for dune fixation, preservation and restoration of ecosystems. Their ability to fix nitrogen from the root symbiosis with bacteria commonly known "rhizobia", improves soil fertility. The effective exploitation of the symbiotic fixation of atmospheric nitrogen to the improvement of agricultural production requires not only selecting the best host cultivar but also requires that the rhizobia population matches and to be characterized very well. In this context, phenotypic and genotypic study of bacteria isolated from several species of legumes tribe Genisteae such as Lupinus sp. and Retama raetam located at several regions of Algeria has been conducted. The phenotypic and genotypic characterization showed that the bacterial isolates studied present a great heterogeneity for selecting strains adapted to highly variable environmental conditions (humid, semi-arid, arid, alkaline or acidic soil, resistant to antibiotics and heavy metals,...). These results can be taken for a possible story and possible application to agricultural scale.

Keywords: Rhizobia, Genisteae, biodiversity, phylogeny, root nodules.

# Enzymatic Screening, Antibacterial Potential and Molecular identification of actinomycetes Isolated from Laghouat region, Algeria

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## Abstract

Actinomycetes play an important role in the decomposition of various organic materials and replenish the supply of nutrients in the soil. They also produce many secondary metabolites possessing antimicrobial properties. The present study was conducted to determine the enzyme- producing and antibacterial potential of soil actinomycetes isolated from an arid soil of Laghouat in the South-East of Algeria. A total of 15 thermophile isolates were recovered from soil sample (50°C as maximum growth temperature). According to morphological and phylogenetic analysis of 16S rRNA gene sequences, these isolates were identified as members of the genus Streptomyces and Micromonospora. Also, they were screened for their ability to produce chitinase, cellulase, amylase and lipase. Screening for the enzyme production was carried out by observing the clear zones surrounding the colonies on agar plates containing specific substrates. The results revealed that 80%, 60% and 93.33% of the isolates produced cellulase, amylase and lipase, respectively, while 53.33% of the isolates were found to produce all the three enzymes. Chitinase production was demonstrated by all the isolates. Screening for antibacterial activity revealed 73.33% of the total isolates inhibited some Gram positive and Gram negative bacteria. The excellent enzymatic potential and antibacterial properties of these strains may find various industrial and pharmaceutical applications, respectively.

Keywords: Actinomycetes, *Streptomyces* sp, *Micromonospora* sp, Antibacterial Activity, Characterization.

# Characterization of phospahte solubilization activity of rhizobacteria strains and plant growth promotion in canola plants

#### Ilhem Saadouli, Raoudha Ferjani, Marwa Cherni, Abdellatif Boudabous and Imene Ouzari Cherif\*.

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# Abstract

Plant Growth Promoting Rhizobacteria (PGPR) have been known to play an essential role in improving crop growth and health. Isolation and selection of microorganisms exhibiting important plant growth promotion features such as phosphate solubilization is a strategic way for developing efficient inoculants especially within poor soil. In fact, Phosphorus is the second important key element after nitrogen as a mineral nutrient for plant growth. In this context, bacterial strains, isolated from date palm rhizosphere, were evaluated for plant growth promotion abilities (phosphate solubilization, cellulose, siderophore, ACC-deaminase, AIA, and NH3 production). Moreover, phosphate solubilization activity including inorganic and organic forms was characterized and examined in different stressful conditions such as salinity, high temperature and basic pH. Two strains of Arthrobacter sp. and Rhizobium giardini exhibited NH3 and IAA production. In addition, Rhizobium giardini was found to produce a high amount of ACC-deaminase (217.05 nmol/mg/h). Phosphate solubilization activity associated with the tested strains decreased in high temperature, whereas the activity was maintained with 10% of NaCl and pH=10 carried on Pikovskaya and phytate specific media. Furthermore, investigated strains were inoculated on Canola plants (Brassica napus L.) to evaluate plant growth promotion efficiency in vivo. In the light of this work, the studied rhizobacteria could be an efficient alternative in phosphate deficient environment for plant inoculation and agriculture sustainability.

Keywords: Phosphate solubilization, Arthrobacter sp., Rhizobium giardini, Plant Growth Promotion and canola.

#### Toward species designation in Frankia genus

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#### Abstract

Filamentous sporangia-forming diazotrophic actinobacteria of the genus Frankia are best known as plant symbionts of dicotyledonous plants, but also found as free-living soil dwellers. Except one not validly named species (Frankia alni), this genus has not yet been described at species level. Analysis of the molecular phylogeny of cultured and uncultured Frankia strains consistently identifies four main clusters regardless of the typing locus used. Cluster 1 includes Frankia strains which associate with Betulaceae, Myricaceae, Casuarinaceae plants (except *Gymnostoma*), while cluster 2 contains and the uncultured Frankia microsymbionts from the Coriariaceae, Datiscaceae, and Rosaceae families as well as Ceanothus of the Rhamnaceae. Frankia strains in cluster 3 form effective nodules on plants from members of the Myricaceae, Rhamnaceae, Elaeagnaceae, and *Gymnostoma* of the Casuarinaceae. Cluster 4 forms а broad group of atypical Frankia strains (non infective and/or non-nitrogen-fixing) that are unable to establish or re-establish an effective association with actinorhizal plants. The current study reports preliminary data gained from genome analysis together with phenotypic array and chemotaxonomy trials performed on a set of strains covering the four *Frankia* clusters.

## Pistacia atlantica roots and fungal endophytes in hyperarid environment

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#### Abstract

Communities of root-associated fungi commonly have been studied under the auspices of arbuscular mycorrhizal fungi (AMF) or ectomycorrhizal fungi. However many studies now, indicate that other groups of endophytic root fungi including dark septate endophytes (DSE) are more abundant in some plants and environments. Several studies have shown the effectiveness of these mycoendophytes to help host plant to overcome severe arid conditions and could improve its growth by mineralizing nitrogen and phosphate components present in the rhizosphere of those poor environments. This is the case of roots Atlas pistachio sampled in hyperarid area (Béni-Ounif; Béchar). Its bleached roots, stained with trypan blue and observed by light microscopy showed the presence of septate hyphae of brown color which traverse the rhizodermis; other follows the sinuosities of intercellular spaces in cortical parenchyma; many mature and immature microsclerotia are also noted within cortical cells. The central cylinder of the observed root is colored blue, demonstrating the presence of nonmycorrhizal fungal structures in the vessels. These could be in (anatomic or systemic) continuity with the same DSE, as shown by several previous works.

# Studies on the physico-chemical properties of the soil samples collected from the rhizosphere of actinorhizal plants associeted microorganisms

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## Abstract

Soil is a vital part of the natural environment, medium of unconsolidated nutrients and materials, forms the life layer of plants and microorganisms. The physico-chemical parameters of soil determine the level of biological activity that can be supported by the soil. The present work has been carried out to study some parameters of soil samples collected from the rhizosphere of actinorhizal plants (*Casuarina equisetifolia, Elaegnus angustifolia*). These trees are used for soil rehabilitation and they are associated with PGPR agents (*Pseudomonas, Frankia*). The soil characterization was carried out for the parameters: pH, conductivity, salinity, humidity, organic material, Carbon (C), Nitrogen (N) and Calcium Carbonate (CaCO<sub>3</sub>). Study of physico-chemical parameters is important for plants growth and soil interaction. It is concluded from the data, the pH scale indicates alkalinity and the soils have *low organic matter* content. The salinity and conductivity of all soil samples is found to be very less while the rate of Calcium Carbonate are too high. These parameters affect the microbial population in soil. However, it was observed that the soil is rich in Nitrogen and Carbon which are essential nutrients for the activity and the development of microorganisms.

Key words: Actinorhizal plants, Rhizosphere, microorganisms, physico-chemical properties.

# Identification of novel antibacterial compounds using virtual screening

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# Abstract

Bacterial drug resistance is a worrying public health problem and there is an urgent need for research and development to provide new antibacterial molecules. Peptide deformylase is essential in variety of pathogenic bacteria, but it is not required for cytoplasmic protein synthesis in eukaryotes and is therefore an interesting potential target for antibacterial agents. The aim of this study was to identify novel and more efficient peptide deformylase inhibitors. Virtual screening using molecular docking is a new branch of medicinal chemistry, which aims to predict the binding mode and binding affinity of a complex protein-ligand. It plays an important role in the rational design of drugs. The binding modes of a series of more than 1500 compounds selected from Zinc database with four human pathogenic bacteria peptides deformylase have been investigated by one of the most established molecular docking program, FlexX version 2.1.8, 2014. The molecular docking approach using the catalytic site of peptide deformylase was successful in identifying different compounds with inhibitory activity. The best result was obtained for the compound pyridine-2,6-dicarbohydroxamic-acid, it showed better binding energy compared to the other compounds. This inhibitor showed excellent *in silico* activity against *Staphylococcus aureus* ( $\Delta G = -35.7764$  kj/mol), moderate against Streptococcus pneumoniae, Escherichia coli and Pseudomonas aeruginosa (-32.4418; -32.2572 and -31.8719 kj/mol respectively). The interactions that govern the complexes deformylase- compound pyridine-2,6-dicarbohydroxamic-acid stability peptides are hydrophobic type and hydrogen bonds. The verification of these results by in vitro tests is underway.

Keywords: Bacterial drug resistance, peptide deformylase, Virtual screening, molecular docking, FlexX,

# Demonstration of foliar mycoendopytes in *Peganum harmala* of dayete Aïat (Laghouat)

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## Abstract

Higher plants are a true ecological niche, a huge potential reservoir of microbial diversity. The mycoendophytes represent an important component of microbial diversity, and are affected by the diversity of plant communities and their structure. They are present in virtually all organs of a given host plant. We were interested in our study to a medicinal plant from the Laghouat region, widespread in the steppe zones: *Peganum harmala* known as the harmel or harmal el sahari rich in alkaloids and volatile substances. We have highlighted the presence of mycoendophytes in the leaves. The observation under an optical microscope shows the presence of very small colored stomata, as trichomes. The parenchyma cells and vessels appear brown and purplish blue. The mycoendophytes can improve tolerance to abiotic and biotic stress. They are the source as well as the plant secondary metabolites with useful biological activities.

Keywords: mycoendophytes, adaptation, Laghouat, Peganum harmala, leaves.

# In vitro study of the antibacterial effect of *Thymus capitatus*, thymol and carvacrol

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## Abstract

Recently, several studies have highlighted the different biological activities of medicinal and aromatic plants, for their powerful antifungal, antibacterial and antioxidant effects. Thus, essential oils have been used for food preservation, industrial perfumes, cosmetics, pharmaceutical and food industries. This study was carried out to evaluate antibacterial activities of Thymus capitatus from Tunisia, Thymol and carvacrol. The antimicrobial activity was also tested against five pathogenic bacteria: *Escherichia coli, Staphylococcus aureus, Salmonella typhymurium, Pseudomonas aeruginosa* and *Bacillus cereus* using the disc-diffusion assay. The inhibition zones diameters for five bacterial which were sensitive to the essential oil, thymol and carvacrol, were in the range of 22,5–45,5 mm. *Thymus* essential oils and carvacrol showed higher bactericidal effect than thymol (17mm) on *Staphylococcus aureus* (31,5mm-30mm). The highest activity was found against *Bacillus cereus* with diameter 45mm and 45mm for *Thymus* and carvacrol successively. The minimum inhibitory concentration (CMI) and minimum bactericidal concentration (CMB) of tested essential oil was determined using agar dilution method. This study showed that carvacrol may be replaced *Thymus* essential oils thanks it important inhibitory power.

# Antagonistic interactions between halophilic archaea in hypersalin environment and characterization of halocin

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# Abstract

Hypersalin environments with NaCl concentrations approaching saturation are often populated by dense microbial communities. Red halophilic Archaea of the family Halobacteriaceae dominate in such environments. The application of molecular biological techniques, in particular the use of approaches based on the characterization of ribosomal RNA sequences, has greatly contributed to our understanding of the community structure of halophilic Archaea in hypersaline ecosystems. Halocins are bacteriocin-like proteins or peptides produced by many species of the family Halobacteriaceae. Halocin production may be expected to be of considerable ecological advantage, as the ability to compete for nutrients and other resources may be enhanced by excreting halocins. However, it has never yet been proven that halocins are indeed excreted by natural communities of halophilic Archaea in concentrations sufficient to inhibit the development of competing strains, thus substantiating their ecological role. In this work, we studied the microbiological, taxonomic, biochemical and spectrometric aspects of antimicrobial activities of new strains of extreme halophilic Archaea isolated from Sebkha Sidi Ameur in the Wilaya M'Sila. The active molecule was synthesized on a large scale through the Platotex. The latter was used for the first time for this type of bacteria to produce large amounts of inhibitory substances for purification studies and characterizations. A biologically active molecule pure was obtained and its amino acid sequence N-terminal and its molecular weight was determined.

Keywords : Halophilic Archaea, Halocin, Hypersalin environments, Sebkha Sidi Ameur.

# Phenotypic Characterization of tolerant to heavy metals rhizobia isolated from contaminated soils in Northeast Algeria

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# Abstract

Twelve rhizobia strains isolated from root nodules of leguminous in Northest of Algeria phylogenetically grouped with *Bradyrhizobium Alpha proteobacteria* were characterized phenotypically, resistance rates and sensitivity of its stem heavy metals were determined in to study their impact on rhizobial growth. Isolated rhizobial strains are Gram-negative bacteria, sticks, on YMA medium the colonies are very small, mucosa, opaque with generation time greater than 6 hours. All strains are sensitive to copper; a low resistance is denoted in presence of cadmium which is 8.33%. Considerable resistance is found with chromium (66.66%) and lead (91.66%). However, all strains are resistant to zinc and mercury (100%). The test of resistance and sensitivity to heavy metals carried on liquid medium confirmed the results found earlier, and showed that most of strains grow in the presence of low concentrations and showed an inverse proportion to the increase in growth with heavy metal concentrations

Keywords: Rhizobia, legume, heavy metals, impact, resistance.

# Preview of bacterial community in the nodules of the actinorhizal tree *Coriaria myrtifolia*

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## Abstract

The diversity of plant-symbiotic nitrogen-fixing *Frankia* strains and its associated bacterial community from *Coriaria myrtifolia* isolated from Algeria, was investigated by terminal restriction fragment length polymorphisms (T-RFLP) technique. Whole DNA from field-collected nodules was amplified by PCR with marked primers targeting genes for the small-subunit rRNA (16S ribosomal DNA). AluI digestion of the PCR products allowed the identification of T-RF haplotypes among the endophytic and epiphytic *Coriaria myrtifolia* root nodules bacteria. T-RFLP profile was converted to binary data using size, height and area. Analyses by a web-based resource, Microbial Community Analysis (MiCA) to compare different communities based on similarity or distance measures. Results of this approach showed that *Coriaria myrtifolia* root nodules contain cluster2 *Frankia* as a major symbiotic endophytic actinobacteria with the presence of other genera such as *Micromonospora, Nocardia, Blastococcus, Rhizobium, Achromobacter* and *Corynebacterium*.

# Aspergillus and Epicoccum: foliar mycoendophytes of Atlas pistachio of dayate aiat (Laghouat, Algeria)

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# Abstract

The plants are a major source of bioactive compounds. These are also colonized by fungi called mycoendophytes. They are able to colonize the internal healthy plant tissues, without causing damage. The relationship with the plant is mutual. Indeed, these fungi are capable of synthesizing the benefit of the plant defensive molecules against various pathogens in the environment. Our study focused at the Atlas pistachio (*Pistacia atlantica* Desf.) known for its ecological amplitude and remarkable plasticity. Our sample was done on the Atlas pistachio of dayate Aiat, wilaya of Laghouat. The leaves were collected in the month of April 2013 in ten healthy individuals. Twenty sheets per tree were chosen for culturing on PDA medium Incubation was done at ambiant temperature for 2 months. This is followed by a morphological identification (macroscopic and microscopic). Several isolates of endophytic fungi were collected from 400 samples cultured. Two genera are dominant: *Aspergillus* and *Epicoccum. Aspergillus* is known for its antimicrobial activity. *Epicoccum* represents a biological control agent, currently being developed commercially because of its ability to produce secondary metabolites with antioxidant and antimicrobial activity.

Keywords: Pistacia atlantica L., Laghouat, Algeria, mycoendophytes, leaf.

# Phylogenetic and plant growth promoting characteristics of *Bacillus* isolated from wheat rhizosphere

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## Abstract

The rhizobacteria that promote the growth of plants, can have a positive effect on the productivity of crops especially in stress conditions. Among Plant Growth Promoting Rhizobacteria (PGPR) cluster, Bacillus is one of the most potential genera due to their spore forming ability, thereby increasing the adaptation of Bacillus strain to commercial formulation and field application. The Bacillus by their intrinsic properties have several mechanisms having beneficial effects to plants. Thirty five strains of Bacillus isolated from the rhizosphere of wheat from three different soils in arid and semi-arid areas of Algeria were tested for their effectiveness in improving plant growth. The ability of strains was evaluated by determining the activities of biofertilisation (solubilization of phosphates), biostimulation (indole acetic acid production: IAA) and biocontrol (cyanhydric acid: HCN, sidérophores, 2,3 butanediol production and antifungal activity). 78% had the ability to solubilize phosphates at rates of 16,65  $\mu$ g / ml for strain D13, 15,60  $\mu$ g /ml for D7 and 15,05  $\mu$ g / ml for D6. These strains were the most successful and were isolated from arid and alkaline soils. The highest concentrations of IAA were produced by strains D4 and D7 to values ranging from 10 to 19µg/ ml. All strains inhibited at least one fungal strain tested and 75% had activity against three fungi and more. More than half of *Bacillus* strains produced 2.3- butanediol but a single strain produced HCN. Only three strains (B25, D11 and BA11) were the most efficient in the production of siderophores. Also four strains (B21, D4, B10 and B25) possessed the ACC deaminase and were considered regulators of stress. Phylogenetic diversity of strains was analysed by 16S rDNA sequencing. The identification results revealed that all of them were similar with Bacillus sp. cluster and was separately divided into five groups. The majority of strains (n=28) were assigned to the species *B. thuringiensis* and *B. subtilis*. The Bacillus species isolated in this study showing PGP abilities have the potential to be used as plant growth promoter rhizobacteria.

Key words: Bacilli, Plant growth promotion, 16S rRNA.

# Response of inoculation by rhizobacteria solubilizing phosphates *Bacillus* sp on growth of durum wheat under salt stress

#### Hafsa Cherif-Silini\*, Bilal Yahiaoui and Allaoua Silini

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## Abstract

Among the various environmental stresses, soil salinity limits the growth and crop production in many parts of the world, especially in arid and semi-arid areas. The use of phosphate solubilizing microorganisms (MSP) as inoculant increases the availability of this element for plants and crop yields. Among these MSP, Bacillus show significant activity for this element. Bacillus strains (n=41) were isolated from rhizosphere soil. They were tested on NBRIP solid medium and the amount of phosphate released was measured on liquid medium NBRIP (10jours/30°C). The majority of strains (75%) solubilize phosphates at rates ranging from 2.89 to 16.65µg/ml P2O5. The strain Bacillus sp. SCH1 having the highest rate of solubilization and with other activities was identified by sequencing the 16S rRNA gene. Inoculation of seeds of durum wheat Triticum durum var. Waha by Bacillus sp. SCH1 is performed in hydroponic medium under salt stress (0, 100 and 200 mM of NaCl). The results show that inoculation significantly increased the weight of the aerial part and root of the plant. Under salt stress, the increase of the chlorophyll content in presence of Bacillus sp. SCH1 is remarkable, while the accumulation of proline and potassium loss are less pronounced. This study reveals that *Bacillus* sp. SCH1 could be an effective biofertilizer for the development of agriculture in arid saline areas.

Key words: inoculation, Bacillus sp, Triticum durum, NaCl.

# Screening of actives bio-molecules of plant growth promoting rhizobacteria isolated from Algerian arid soils

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## Abstract

The use of microbial technology in agriculture extends currently relatively quickly with the identification of new more efficient bacterial strains in the growth of plants. The rhizobacteria that promote the growth of plants (PGPR, plant growth promoting rhizobacteria) can have a positive effect on the productivity of crops especially when subjected to salt stress. Indeed, the salinity considerably affects the crop yield. Inoculating PGPR can mitigate the effects of this type of stress by direct or indirect mechanisms. To select effective strains and having multiple activities, a total of 22 bacterial strains are isolated from different arid soils. These strains are characterized by their ability to fix nitrogen (nitrogen-free medium culture). They are also tested in vitro for their activities of improvement of plant growth such as the production of the indole acetic acid (IAA) of hydrogen cyanide (HCN), of the siderophore, ammonia (NH<sub>3</sub>), of solubilization of phosphates and antifungal activity. More than 70% of the strains produce NH<sub>3</sub>, 54% solubilized more P<sub>2</sub>O<sub>5</sub> 10µg/ml and the same rate and has antifungal activity against to three fungal strains. However, 36% produce more 80µg/ml of IAA, 27% produce siderophores and only two strains produced of HCN. The production of IAA of eight (08) strains is evaluated in the presence of 100, 200 and 300 mM NaCl. The remaining active strains with high concentration of NaCl would be the most effective and can be considered as biofertilizers for improving plant growth in areas affected by salinity.

Keywords: PGPR, arid soil, active biomolecules, salinity.

# Growing varieties durum wheat (*Triticum durum*) in response to the effect of osmolytes and inoculation by *Azotobacter chroococcum* under salt stress

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# Abstract

This study was conducted to determine the effect of plant growth promoting rhizobacteria *Azotobacter chroococcum* AZ6 isolated from arid soil and osmolytes such as glycine betaine (GB) or proline (P) on the growth of durum wheat varieties under salinity stress. Inoculation by *Azotobacter chroococcum* AZ6 in the presence or absence of P (5mM) or GB (5mM) reduced substantially the effect of salt stress on plant growth parameters such as root length, plant height, fresh shoot and root weight and dry shoot and root weight. The differences between the two varieties were low but with a fresh and dry weight higher in Waha. The rate of Na<sup>+</sup> accumulation in the roots and the shoots was important to 100 mM and increased at 200 mM. The K<sup>+</sup> concentration and chlorophyll content decreased but proline and amino acid contents enhanced with increasing salinity. Treatment by inoculation in the presence or absence of osmolytes improved the chlorophyll (*a* and total) and the K<sup>+</sup> concentrations and reduced intracellular proline accumulation and amino acids contents. Also, as result, the use of *Azotobacter chroococcum* AZ6 and osmolytes treatment may provide a means of improving tolerance of durum wheat to salt stress.

Keywords: Durum wheat, salinity, osmolytes, Azotobacter chroococcum,

# The role of natural compatible solutes from *Ulva lactuca*, *Enteromorpha intestinalis* and *Opuntia ficus-indica* in the halotolerance of diazotrophic PGPR and the germination of wheat seeds under salt stress.

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## Abstract

High salinity affects about 20 % of the irrigated agricultural lands being a major threat to agriculture. Diazotrophic bacteria are free-living nitrogen fixers; they are largely inhibited by salt stress. 70 diazotrophic bacterial strains were isolated on the N-free Jensen's medium. The best strains were selected based on their ability to produce enzymes of agricultural interest (protease, lipase, amylase etc...), to synthesize the auxin Indole Acetic Acid (IAA), siderophores and to solubilize tricalcium phosphate. Four strains (BEA4, BEC9, BOA4, and SEB9) were selected and identified as Flavobacterium johnsoniae, Pseudomonas putida, Achromobacter xylosoxidans and Azotobacter chroococcum, respectively. They produce the searched enzymes, solubilize inorganic phosphate, express high amounts of siderophores and synthesize IAA: [76.31 µg/ml] for BEA4, [32.24 µg/ml] for SEB9. The four strains were grown under different NaCl concentration without/with: Glycine Betaine (GB) [1 mM], extracts from Ulva lactuca (UL) and Enteromorpha intestinalis (EI) and Opuntia ficus-indica (OFI) [1%]. Wheat seed germination was tested under 0, 100, 200, 300 mM of NaCl in presence/absence of GB and extracts from UL, EI and OFI. The aforementioned extracts remarkably promote bacterial growth under salt stress compared to the results obtained with the synthetic osmoprotectant GB. The use of natural osmoprotectants and PGPR strains would constitute an efficient alternative to chemical fertilizers under salt stress.

Keywords: PGPR, Salt stress, Diazotrophic bacteria, osmoprotectant.

# Session 04

# **Microbial Biodiversity**

# Keynote 7

# Culturomics, improved media prediction and phylogenomics in Microbial Ecology

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# Abstract

The availability of -omics techniques for the analysis of organisms initiated a dramatic change in microbiology during the last two decades. Sequencing of DNA, RNA and proteins was rather expensive in the early days when the -omics technologies were developed. The dramatic reduction in the price level during the last few years combined with the miniaturisation of the tools and availability ofeasy to usekits made -omics technologies affordable forordinary research laboratories and allows researchers to gain a more holistic view on microorganisms. This progress in analysis techniques is complemented by good progress in novelhigh-throughput cultivation techniques and innovative culture conditions that significantly increased the fraction of viable microbes that can be identified and analysed following on cultivation. Thereby not only the diversity of the cultivatable fraction of the diversity can be assessed to a much higher degree, but also allows for a better description of non-cultivatable fraction of the remaining microbiomes viametagenomics and metatranscriptomics. Together the improved techniques will inevitably lead to shifts in the way microbial cultures will be analysed and looked upon and eventually to improved standards in the description of novel species.

# Keynote 8

# Promises of next generation sequencing for microbial ecology

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# Abstract

Recent advances in molecular biology have resulted in the application of next-generation sequencing technologies (NGS) to the field of microbial ecology. This talk using specific targeted applications of NGS to classical microbial ecology studies mainly in the field of plant pathogen interactions aims to examine the strengths and weaknesses of different NGS protocols, including depth, ease of bioinformatic analysis, throughput and cost-effectiveness. It also intends to highlight diverse new findings using these technologies such as the rare biosphere and the link between communities biodiversity and ecosystem function. Beyond PCR biases that are widely recognized, we will focus on other unsuspected biases that may arise from the sequencing process itself or from post-sequencing quality control algorithms and databases and their impacts on downstream analyses. Whole-genome sequencing, targeted resequencing, RNA expression profiling and transcriptomic studies and their transforming potential to the field of microbial ecology will also be discussed. We suggest that the efficient use of NGS technologies will help to advance the field of microbial ecology, and importantly, improve our understanding of the role of microorganisms in their different environments.

#### Keynote 9

#### The story of Actinobacteria revisited in the light of Bioinformatics

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#### Abstract

Actinobacteria are GC rich gram positive bacteria having adapted to contrasted ecological niches as free living microorganisms or plant/animal pathogens or symbionts. While, Mycobacterium contains mostly pathogens that cause a variety of mammalian diseases like leprosy and tuberculosis, Streptomyces is mostly a soil microbe known for secondary metabolites production. Frankia establishes a root symbiosis with dicotyledonous pioneer plants and is a nitrogen-fixer Geodermatophili are known extremophiles. Thus, they have to take up some special measures to tackle such diversed ecological circumstances. The phylogeny of the class Actinobacteria remains controversial, essentially because it is very sensitive to the choice of dataset and phylogenetic methods. A number of phylogenetic trees were constructed from different genes and proteins. The best tree, with the lowest number of lateral transfers, was the one based on the concatenation of 54 proteins. In that tree, the orders Frankiales and Micrococcales were not. It is thus proposed that the order Frankiales, be split into Frankiales ord. nov., Geodermatophilales ord. nov., Acidothermales ord. nov. and Nakamurellales ord. nov. The order Micrococcales should also be split into Micrococcales, Cellulomonales and Brachybacteriales. A comparative genome analysis of the secretomes of symbionts and pathogens allows a thorough investigation of selective pressures shaping their evolution. It was found that secreted proteins as a whole have a stronger purifying evolutionary rate (non-synonymous to synonymous substitutions or Ka/Ks ratio) than the nonsecretory proteins in most of the studied genomes. This difference becomes statistically significant in cases involving obligate symbionts and pathogens. Even within the secretory proteins, the signal peptide part has a higher Ka/Ks ratio than the mature part. The higher evolutionary rate of signal peptide part compared to mature protein provides an indication that signal peptide parts may be under relaxed purifying selection, indicative of the signal peptides not being secreted into host cells. Functional domains are semi-autonomous parts of proteins which play a significant role in signal transduction. The Per-Arnt-Sim (PAS) domain functions as signal-sensor in two-component systems of several bacteria. This domain exhibits large sequence diversity and makes associations with co-domains to modulate their function. From 3D structure based phylogeny tree it was hypothesized that the structure of PAS domain may determine its associating co-domains as well as biological network or viceversa and also may help in niche adaptation in selected actinobacteria.

# **ORAL COMMUNICATIONS**

## **MB-01**

#### Blastococcus capsiensis sp. nov., isolated from an archaeological Roman swimming pool

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## Abstract

A novel Gram-positive actinobacterium designated BMG 804<sup>T</sup> was isolated from an archaeological Roman swimming pool located in Gafsa, Tunisia. The strain produced dry bright orange colonies and grew well at 30°C and pH 6.0-8.0. Chemotaxonomically, it contained *meso*-diaminopimelic acid in the cell wall. The cell-wall sugars contained glucose, ribose. Polar lipids were diphosphatidylglycerol, rhamnose and present phosphatidylethanolamine, phosphatidylcholine, phosphatidylinositol, glycolipid and two unidentified phospholipids. MK-9(H<sub>4</sub>) was the predominant menaquinone. The fatty acid profile contained major amounts (>5 %) of  $C_{17:1}\omega 8c$ ,  $C_{16:1}\omega 7c$ , iso- $C_{15:0}$ , iso- $C_{16:0}$  and iso-H-C<sub>16:1</sub>. The 16S rRNA gene sequence of BMG 804<sup>T</sup> showed 98.9 % as highest sequence identity with *Blastococcus saxobsidens* DSM 44509<sup>T</sup>. DNA–DNA hybridization between strain BMG 804<sup>T</sup> and *B. saxobsidens* DSM 44509<sup>T</sup> was  $48.6 \pm 6.6\%$ . The G+C content of the DNA was 73.7 mol%. On the basis of the phenotypic and genotypic characteristics, including low DNA –DNA hybridization results, strain BMG  $804^{T}$  (= DSM  $46835^{T}$  = CECT  $8876^{T}$ ) is proposed as the type strain of a novel species *Blastococcus capsiensis* sp. nov.

## Identification of aerobic spore forming and non-spore-forming bacteria isolated from Tunisian milk

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#### Abstract

Milk is highly nutritious beverage, that's why it is an especially suitable growth medium for various bacteria. These microorganisms may cause several economic losses to the producers by causing spoilage of the dairy products. In order to preserve the quality of drinking milk and maintains its freshness, it is important to determine the reasons responsible of organoleptic defects. Spoilage of milk is caused by different mechanisms, and at different moments in the marketing chain from farm to consumer. So it will be very interesting to have an idea about these organisms, in order to minimize spoilage of UHT milk. In order to get an idea about their distribution in the different types of milk, we carried out the isolation of aerobic spore-forming and non spore-forming bacteria using a microbiological screening throughout the chain production of UHT milk in Tunisia. These isolates were identified phenotypic by studying cultural, morphological, biochemical characteristics and genotypic using the 16S ADN gene. Different bacteria from eight genera have been isolated, both grampositive «Bacillus spp, Staphylococcus spp, Microbacterium spp... » and gram-negative «Pseudomonas, E.coli ...». The current study revealed that the predominant organisms were the genus *Bacillus* and *Microbacterium* whose are described as heat resistant species. Despite the effect of ultra high temperature treatment, the obtained results show the persistence of a potential risk of foodborne illness in UHT milk.

Key words: Milk, Spore-forming bacteria, Non spore-forming bacteria, Bacillus

#### Investigation of Nattrassia mangiferae Isolated from Date Palm on different Hosts

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#### Abstract

The present work is concerned with the study of fungus *Nattrassia mangiferae* Nattrassas a causal organism of sooty stem canker disease and leaf die back of date palm (Phoenix dactyliferaL) in the Northern state of the Sudan.Symptoms of N. mangiferae in date palm first appear as chlorosis on the tip of the leaf followed by necrosis and dryness, trees wilt and dieback, black sooty canker layers of spores are revealed when the infected part wasremoved from infected areas. The vegetative mycelium grew readily and formed a whitish mat in colour within 2-3 days, and then turn to blackish in colour .The colonies of N.mangiferae effused dark blackish brown to black, Hyphae mid to dark brown, smooth 2-8 µ thick, conidia (arthroconidia) mid to dark brown, smooth, mostly aseptate but occasionally with one or more very dark transverse septa, cylindrical 4-17 x 3-8  $\mu$ , or spherical to sub spherical and 5-11 $\mu$ in diameter. Physiological studies of the fungus include temperature, suitable medium, and pH and carbon source. The maximum temperature for the fungalgrowth was 35° C.The PDA medium was found to be the best medium for the growth compared to other tested media. The best carbon source for the growth of the fungus was dextrose. The hydrogen ion concentration at level pH 5 gave the maximum growth of the fungus colony. Pathogenicity experiment proved that the symptoms of infection were produced in artificial inoculation similar to those caused by N. mangiferaein nature. Root inoculation was more successful as compared to stem soil inoculations. The host range of *N.mangiferae* is extremely wide and it includes plants belonging to different plant families' e.g. Citrus spp., Guava, Ficusspp, Mahogany, Albizia, Acacia nilotica, Ficusnitida, Ficubengamina, Eucalyptus camaldulensis, Khayasengalenesis, Acciaseval, Acacia sengal, Termnaliacatappa, Albizialebbek, Adonsoniadigitata, Cassia nodosa, and Psidium spp.

Keywords: date palm (Phoenix dactyliferaL), sooty canker, Nattrassia mangiferae.

# Flow cytometry approach for studying the interaction between *Bacillus mojavensis* and *Alternaria alternata*

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#### Abstract

Tomato (*Lycopersicon esculentum* Mill) is one of the most important vegetable consumed in Algeria. Tomato crops are often infected by *Alternaria alternata*, which causes early blight disease. Chemical pesticides are intensively used to protect this plant, which lead to environment pollution that might endanger animal and human health. The main objective of this study is to select potential biocontrol agents from arid soil as an alternative to chemical products. The phytopathogenic fungus which was isolated from infested tomato leaves, stems and fruits cultured in (Constantine-Algeria), was identified as *Alternaria cf. alternata*. Thirty five bacteria isolates were obtained from arid soil in the south of Algeria. Three of the isolates inhibited the growth of *A. alternata*. However, the most potent isolate E1B3 reached a 75% inhibition rate. The molecular identification of this isolate showed that it was closely related to *Bacillus mojavensis* (KC977492). This strain does not produce chitinase, but does produce lipase, protease and lipopeptides. The interaction between *A. alternata* and *B. mojavensis* was investigated in the first time in this work by flow cytometric analysis. To conclude, *B. mojavensis* strain was antagonistic to *A. alternata* which could possibly be exploited as a biopesticide in tomato crops management.

Keywords: Tomato, biocontrol, *Bacillus mojavensis*, early blight, *Alternaria alternata*, and Flow cytometry.

#### **MB-05**

## Preview of salt tolerant and phosphate solubilizer bacteria from date palm rizobiome and their plant growth promotion potentialities

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#### Abstract

Salt stress is one of the most serious environmental limitations causing substantial crop losses, it's in steadily increasing particularly in arid and semi-arid areas. Salt stress was shown to cause reduction in phosphate accumulation in plants, which may develop P-deficiency symptoms. Bacteria living in association with plants have been shown to play an essential role in increasing the plant growth, through direct and indirect effect. In this context, our study intended to (i) study the diversity of salt tolerant bacteria solubilizing phosphate associated with date palm rhizobiome, (ii) select the most potent strains for plant amendment experiments, through the screening of some plant growth promotion (PGP) traits and to (iii) in vivo evaluate the efficiency of selected strains on the plant growth promotion under salt stress condition. The diversity of date palm microbiome was assessed according to culture dependent approach. A collection of 202 isolates was obtained using Pikovskay phosphate medium added with an increasing concentration of NaCl (10%, 13%, 16% and 20%). A wide genetic diversity was recorded based on 16S-23S rRNA internal transcribed spacer ITS and 16S rRNA gene sequences. Bacterial strains were clustered into Firmicutes (62%), y-Proteobacteria (30%) and Actinobacteria (8%). Two strains belonging to the genus Bacillus and Oceanobacillus, and exhibiting high level of PGP potentialities, were selected and inoculated on tomato plants, irrigated with salt water (100mM and 150mM of NaCl). The result showed that the applied strains displayed significant effects on the recorded plant growth parameters. Selected halotolerant phosphate solubilizing rizobacteria from date palm trees are promising biofertilizers candidates for sustainable crop production in arid lands.

# Phenotypic and genotypic diversity of bacteria isolated from root nodules of Lucerne (*Medicago sativa* L.) cultivated in Algeria.

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#### Abstract

The aim of this study is to determine the biodiversity of strains isolated from nodules of Lucerne (*Medicago sativa* L.) cultivated in soils from various regions of Algeria. A total of 106 strains were characterized on the basis of morphological, cultural and phenotypical properties. Phenotypic characteristics studied included growth rates, colony morphology, symbiotics traits, utilization of different carbon and nitrogen sources, tolerance to different abiotic stress such extremes temperature, salt, pH, drought and resistance to different antibiotics and heavy metals. Polymerase chain reaction reaction (PCR) method and 16S rRNA gene sequencing were used for the genotypic analysis of some isolates selected on the basis of phenotypic study. Numerical analysis of the phenotypic characteristics showed that the 106 strains studied fell into diverse groups reflecting a wide biodiversity. RFLP analysis of 16SrRNA genes revealed an additional heterogeneity and 12 ribotypes were identified. This diversity is not clearly correlated with their geographical origin and soils characteristics (pH and salinity) does not seem to affect this diversity. This is the first report evidencing that Lucerne nodule may be associated with strains such as *Stenotrophomonas maltophila* and *Rhodococcus quinshengii* identified by sequencing of the 16S rRNA.

Keywords: Biodiversity, Phenotypic, Ribotype, Strain, Lucerne.

# POSTERS

# Bacteriological quality of surface water in Northeast Algeria. Case of Oued El-Kebir Ouest (Ben Azzouz, Skikda)

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## Abstract

Oued El-Kebir Ouest is located in Ben Azzouz Township (Skikda city, Northeast Algeria), it plays an important role in the regional economy by providing water for irrigation and feeds several lakes belonging to Guerbes-Sanhadja Wetland complex. However, the discharge of untreated sewage from Ben Azzouz, industrial effluents, breeding, intensive agriculture and the excessive use of pesticides may be major sources of pollution in the study area. The present study aims to the evaluation of the bacterial quality of Oued El-Kebir Ouest. Five sampling locations were selected and total 20 samples were collected over a period of four seasons from winter 2011 to summer 2012. The bacteriological analysis showed that total coliforms (ranged from 2.0 x 10<sup>4</sup> to 9.1 x 10<sup>9</sup> MPN/ml), fecal coliforms (ranged from 2.5 x  $10^4$  to 9.9x  $10^8$  MPN/ml), fecal streptococci (ranged from 1.1 x  $10^4$  to 2.3 x  $10^7$  MPN/ml), sulphite reducing Clostridia and total viable microorganisms counted in the river water samples were found to be greater than the standard WHO limits for agricultural use. Nineteen bacterial isolates were recovered from all samples included pathogenic species: Staphylococcus aureus, Salmonella spp, Shigella spp, Pseudomonas aeruginosa. The study shows that the studied water was heavily bacteriologically polluted and of serious threat to the aquatic biota and public health.

**Keywords:** Oued El-Kebir Ouest, Water quality, bacteriological parameters, fecal contamination, microbial pollution.

# Genetic diversity and inoculum exchange of *Botrytis cinerea* between tomato reenhouses in Northern Algeria

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## Abstract

To estimate the genetic diversity for a better understanding of the spread of *Botrytis cinerea*, we genotyped with nine microsatellite markers 174 isolates collected from four greenhouses during three growing seasons in the region of Bejaia. Four of these isolates were detected as *Botrytis pseudocinerea* according to the allele size at locus Bc6. For all other isolates further studied, all loci were polymorphic, with the mean number of alleles per locus ranging from 2.77 to 5.22. Considerable genetic variability was detected in all subpopulations (D\*>0.87; Hnb>0.40). Based on the standardized index of association analysis, significant but low levels of clonality occurred, not excluding the possibility of recombination (rD= 0.07, P<0.001). A total of 109 haplotypes were characterized among the isolates, few of which were shared between subpopulations. This, together with moderate genetic differentiation among subpopulations according to the geographical origin (0.080<F<sub>ST</sub><0.167), suggested a low level of inoculum exchange among greenhouses and little carry-over of inoculum from one sampling season to the next. The importance of genetic structure of *B. cinerea* populations is discussed and should be taken into consideration for the management of grey mould.

Keywords: Genetic diversity, Grey mould, Inoculum, Exchange, Microsatellite makers

# Characterisation of rhizospheric actinomycetes antagonists of Fusarium spp.

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## Abstract

Plant diseases cause a significant loss, it is 40 % in industrialized countries and over 50 % in developing countries. The rhizosphere is rich in actinobacteria. This bacterial group have strong capability in biocontrol against plant diseases. In this study, isolation of plant pathogenic fungi was conducted on infested vegetable plants and cereal crops. The three phytopathogenic fungi isolated, identified and selected are: F. culmorumTRI-1, F. oxysporumCIT-4, M. nivaleTRI-2.Furthermore, 72 actinobacteria strains were isolated from different rhizospheric soils sampled in the semi-arid region of Algeria. The in vitro antagonism of the actinobacteria isolates was tested against the three phytopathogenic fungi. So, six isolates (Lac1, Lac3, Vic8, Pin10, Pru14 and Pru16) have a chitinolytic activity, and inhibited the growth of all tested fungi, were identified by the study of their morphological and physiological characteristics and phylogenetic analysis sequences of the 16S rRNA gene (1500 bp). Four of them were assigned to the Streptomyces genus. While, the other two isolates Vic8 and Pin10 were classified in the *Nocardiopsis* genus. This study shows the large isolates proportion of Nocardiopis (33%) among with antifungal activity.

**Keywords**: Phytopathogenic fungi, antifungal activity, rhizospher, isolation, *Nocardiopsis*, *Streptomyces*.

# Isolation of bioactive actinomycetes from soil samples collected in water source in the region of Tizi-Ouzou, North Algeria

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## Abstract

Microbial biodiversity in various ecological niches is a reservoir still under-exploited with promising potential in the field of biotechnology. The exploration and exploitation of this biodiversity is therefore of vital importance, particularly in finding therapeutic tools that can respond to major public health issues such as the emergence and spread of multi-resistant bacteria. As part of a study to isolate strains of actinomycetes producing potentially novel antibacterial molecules, ten soil samples were collected near a water source located in the locality of Ain El Hammam, South-East of Tizi-Ouzou district (North Algeria). A total of 27 strains were isolated and screened for antibacterial activity against three reference strains (Escherichia coli ATCC 25922 Pseudomonas aeruginosa, and Staphylococcus aureus ATCC 727853, ATCC 25923) by two agar diffusion methods: the perpendicular streak method and the agar cylinder method. Of 27 isolates, 18 showed an activity toward at least one test bacterium, of which 13 are highly active against Staphylococcus aureus with inhibition diameters between 20 and 27 mm, 4 having an activity against 3 test bacteria and a particularly active one against *Pseudomonas aeruginosa* with a 30 mm inhibition diameter. Five isolates of the most active were selected for identification. The results attest of the wealth of this region in terms of microbial strains of biotechnological interest and seem to be promising for possible applications against multi-resistant bacteria.

**Keywords:** Biodiversity; Actinomycetes; Antibacterial activity; Bioactive compounds; Multiresistant bacteria.

# *Enterobacter biskrae* sp. nov. and *Enterobacter zibanae* sp. nov., two glyphosatedegrading bacteria isolated from soil in Biskra, Algeria

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## Abstract

Degradation of xenobiotic compounds is an important indicator for healthy ecosystems. The application of these compounds generates environmental concern by the potential for unwanted side effects. Soil microorganisms can carry out pesticide degradation and can use the xenobiotic as a source of carbon, energy and other nutrients to promote microbial growth. Glyphosate is the most commonly used herbicide worldwide. Because of concern regarding its toxicity for non-targeted species in soil, finding glyphosate-degrading organisms in soil is of interest. The success of this will depend on isolating bacteria with the ability to degrade glyphosate. Two such rod-shaped, motile, Gram-negative, aerobic catalase positive, oxidase negative and glyphosate-degrading bacteria, designated strain Bisph1 and strain Bisph2, were isolated from one soil specimen collected in Biskra, Algeria. The strains were able to grow in a medium containing glyphosate as sole phosphorus source by enrichment cultures of this soil. Combined 16S rRNA gene and rpoB gene sequence, phenotypic and the Matrix-Assisted Laser-Desorption/Ionization Time Of Flight Mass Spectrometry (MALDI-TOF MS) analyses revealed that Bisph1 and Bisph2 strains belonged to the genus *Enterobacter*. Multilocus sequence analysis of the 16S rRNA, hsp60, rpoB, gyrB and dnaJ genes demonstrated that Bisph1 and Bisph2 formed one separate cluster in the phylogenetic tree of *Enterobacter*. On the basis of the phenotypic characteristics and molecular analyses data we propose that strains Bisph1 and Bisph2 represent two novel species of the genus Enterobacter, for which the names Enterobacter biskrae sp. nov. and Enterobacter zibanae sp. nov. are proposed.

**Keywords:** Glyphosate, *Enterobacter biskrae* sp. nov., *Enterobacter zibanae* sp. nov, Multilocus sequence analysis.

### Reclassification of Acetomicrobium faecale as Caldicoprobacter faecalis comb. nov.

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#### Abstract

Acetomicrobium faecale was isolated from sewage sludge incubated at 72°C. This Gramnegative rod-shaped bacterium shared many physiological characteristics with the type species of the genus Acetomicrobium, A. flavidum which justified its assignment to this genus. However, recent phylogenetic analysis revealed that A. faecale, but not A. flavidum together with Caldicoprobacter spp. formed a separate branch within the order Clostridiales. In this respect, A. flavidum and A. faecale should not be considered at the same genus level. Beside these phylogenetic features, further genetic and chemotaxonomic experiments have been undertaken to ascertain reclassification of A. faecale as a novel species of the genus Caldicoprobacter, Caldicoprobacter faecalis comb. nov.

# Inventory of Copépodes parasites of both fishes Cyprinides (*Barbus callensis* and *Cyprinus carpio*) populating the dam Foum-El-Khanga (Souk-Ahras).

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#### Abstract

Among the major fish ectoparasites, copepods occupies a place favored in the world of the parasitism because of their extraordinary adaptive capacity. These parasites present great interest at three different levels. First, these parasites are able to weaken the fish and interfere negatively in the fish's weight-gain, reproduction, growth. Others copepods reveal pathogens and induce mass mortality. Second, parasitic copepods serve as taxonomic, bio-geographical and even hybridization markers. Third, copepods parasites may be good indicators of water quality. Despite their omnipresence in the living world, and their ecological importance, they are very little known on fish of Algerian freshwater. This work is interested in the study of parasitic copepods population harvested on the gills of 240 fish individuals belonging to two species known for their ecological and economic importance as well, Barbus callensis and Cyprinus carpio harvested in Foum El Khanga dam during the period spanning the months from January 2015 to April 2015 at the rate of 30 individuals for each species per month. The examination of fish gilsl allowed us to collect 182 copepods ectoparasites belonging to seven species: Ergasilus sieboldi (Nordmann, 1832), Ergasilus briani (Markewitsch, 1937), Ergasilus peregrinus (HALLER, 1865), Ergasilus lizae (KROYER, 1863), Paraergasilus brevigiditus (YIN, 1954) Paraergasilus longidigitus (YIN, 1954) and Neoergasilus longispinosus (YIN, 1956). The distribution of the parasite indices in both host species shows that it is the species *Cyprinus carpio* that is most affected by the parasitic copepods. We note, in fact, than in Barbus callensis it is the species Paraergasilus brevidigitus which registers the highest infection rate (19.67%). Moreover, in Cyprinus carpio it is the species Ergasilus sieboldi which present the highest infection rate (58.06%). However, the infestation with parasitic copepods varie from a month to another, we note a spring peak in both host species. The Multivariate statistical analysis shows the influence of the size and microhabitat on the infestation of these two fish species by the collected copepods, we note in fact that sex presents no effect on the parasitic infestation.

**Keywords** : Dam Foum El Khanga (Souk Ahras) ; *Barbus callensis* ; *Cyprinus carpio* ; Parasitic copepods ; parasitic indices

# The spread of carbapenemases in Guelma hospitals - Algeria

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### Abstract

The spread of a nosocomial outbreak often originates from cross-contamination and the most common means of pathogen transference occurs between the hands of healthcare professionals, hospital equipment and patients. However, drug-resistant bacteria cause many of these nosocomial infections, with an increasing predominance of imipenem resistant Gramnegative organisms. The study included imipenem-resistant Acinetobacter baumannii isolated in 2014 (January to December) from patients hospitalized for at least 48 hours (urine, blood, pus...) and from environmental samples (surfaces and adjacent equipment of nine wards: emergency, operating room, pediatric, gynecology and neonatal, general surgery, general medicine, infectious diseases, hemodialysis and phthisiology) in Ibn-Zohr and El-Hakim Okbi hospitals in Guelma, Algeria. The isolates were identified by microbiological methods and confirmed by MALDI-TOF MS. Antibiotic susceptibility was performed using disk diffusion and Etest methods. Carbapenemase activity was detected via microbiological tests (MHT, MCNP et EDTA). Carbapenem-resistance determinants and ESBLs encoding genes were search for by PCR and sequencing. A total of 10 imipenem-resistant A. baumannii isolated from clinical and surfaces specimens were screened, 7 of them harboring  $bla_{NDM-1}$  and 3 isolates harboring *bla*<sub>OXA-23</sub>.

Keywords: A. baumannii, imipenem-resistant, carbapenemases, environnement, nosocomial infections.

# Chromatographic separations of secondary metabolites of two strains of *Aspergillus fumigatus*.

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# Abstract

Molds can have very extended biotechnological applications by their ability to conquer natural substrates. Currently, molds are increasingly used in the production of primary metabolites and secondary metabolites such as antibiotics, mycotoxins, among these molds we can mention: *Aspergillus fumigatus* which secretes many secondary metabolites. This study involves the extraction of secondary metabolites of two strains of *Aspergillus fumigatus* isolated from the grains of wheat provided by AIOC (Algerian Interprofessional Office for Cereals) of Constantine and the other from an agricultural soil of the extreme zone (Laghouat). To study the production of secondary metabolites by *Aspergillus fumigatus*, these strains were cultured on Czapeck Dox medium supplemented with yeast extract. The extraction was accomplished with organic solvents, their separation was made on the thin layer chromatography (TLC) with visualization by UV radiation (365 nm and 254 nm) and high performance liquid chromatography (HPLC). Analysis of crude extracts of the two strains of *Aspergillus fumigatus* by TLC showed that the two strains secrete 12 metabolites while analysis by HPLC; 27 metabolites were detected from the strain isolated from the soil and 36 metabolites from the isolated strain of wheat seeds.

Keywords: secondary metabolites, Aspergillus fumigatus, TLC, HPLC.

# Isolation and identification of four *Bacillus* sp. strains from western Algerian soil and their α-amylase production monitoring

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# Abstract

Alpha-amylases (EC 3.2.1.1) are digestive enzymes which hydrolyze glycosidic bonds in starch to ultimately obtain maltose molecules. These enzymes represent the biggest part of the enzyme market and are widely used in diverse industries *ie*: paper, detergent, food, textile, pharmaceutical and biofuel. The first source of  $\alpha$ -amylases is microorganisms due to their availability and cheapness of production and purification process. For commercial applications  $\alpha$ -amylase is mainly derived from the bacterial genus *Bacillus*. In this work four *Bacillus sp.* strains were isolated from western Algerian soil in different area then they were identified using 16S rDNA gene sequencing. Alpha amylase production and bacterial growth were monitored for each strain. The identified strains 16S rDNA gene sequences were submitted to Genbank as: *B.subtilis; B.mojavensis* and two *Bacillus sp.* Amylase production monitoring showed good amylase yields and the production started generally at the beginning of the growth stationary phase. The four strains showed high amylase activity at temperatures higher than 60°C.

**Keywords:** *Bacillus*, identification, α-amylase, production.

# Optimization by response surface methodology of culture parameters for bioactive metabolites production against *Candida albicans* by river sediment *Streptomyces* sp. SRC3

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# Abstract

The intensive use and misuse of antibiotics has built up resistance in most human pathogens against antibiotics. Microbial natural products, particularly those produced by actinomycetes are a valuable source of various clinically important compounds. The following investigations were undertaken for the purpose of finding the optimal culture conditions for antibiotic production by the strain producer Streptomyces sp. SRC3, using Response Surface Methodology (RSM). Thirty nine actinobacterial strains were isolated from Ziama river sediment (jijel region) and screened for antagonistic activity against various bacterial and yeast pathogens. The active antibiotic producer SRC3 was identified as Streptomyces sp. using morphological analysis. Plackett-Burman design (PBD) was employed to screen the influential media components affecting the antimicrobial compound production against C. albicans. Subsequently, statistical optimization of selected medium components was performed by employing the response surface methodology (RSM) with central composite design (CCD). The obtained results from PBD showed that the factors influencing the antimicrobial activity are KCl, MgSO<sub>4</sub>, 7 H<sub>2</sub>O and incubation time. The optimal values of the selected factors were determined through the plot isoresponse curves giving: KCl 0.51 g/L, MgSO<sub>4</sub> 7 H<sub>2</sub>O: 0.50 g/L and incubation time : 5.56 days. This study points the success of statistical model in developing an optimized production media for enhanced antimicrobial compound production by Streptomyces sp. SRC3.

Keywords: Optimization, Response Surface Methodology, River sediments, *Streptomyces*, antibiotics.

# Diversity of thermophilic Bacillus species in two Algerian terrestrial hot springs

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# Abstract

Thermophilic and hyperthermophilic microorganisms inhabiting hot environments such as terrestrial hot springs have interesting molecular adaptation capacities and constitute an important source of bioactive molecules emerging from unique molecular and biochemical mechanisms. In this work, we investigate the diversity of thermophilic, aerobic *Bacillus* strains isolated from water and sediment samples taken from Hammam Ouled Ali and Hammam Debagh hot springs, located in the region of Guelma, Algeria. Phenotypic characterization allowed to obtain some morphological, biochemical and physiological proprieties of these microorganisms. The majority of the strains has shown some interesting proteolytic and/or amylolytic activities. Restriction fragment length polymorphism (RFLP) and phylogenetic analysis of 16S rRNA gene sequences revealed that the bacterial isolates displayed close relationships to several bacterial species of *Bacillus* genera. They were phylogenetically affiliated to the phylum *Firmicutes* and belonged to the genera: *Bacillus*, *Geobacillus, Anoxybacillus, Brevibacillus* and *Thermoactinomyces*.

Keywords: Thermophilic, Bacillus, hot spring, Algeria.

# Diversity of Frankia microsymbionts of Coriaria myrtifolia in Northern Africa

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# Abstract

Diversity of Frankia microsymbionts of Coriaria myrtifolia was studied throughout Northern African area (Morocco, Algeria and Tunisia). DNA extracted from root nodules collected from natural stands and from seedling trapping bioassays was used for PCR-amplification and sequencing of glutamine synthetase (glnA) gene. Two major clades occur in very different geographical and ecological areas and are associated to different host plant species. The first clade includes C. myrtifolia microsymbionts collected from natural stand in Algeria and by plant tapping bioassays in Tunisia together with microsymbionts of C. myrtifolia, C. nepalensis and C. mycrophilla collected respectively from France, Pakistan and Mexico. The second clade was made up by C. myrtifolia microsymbionts collected from Algeria and Morocco and was also detected in France and Japan in association respectively with C. myrtifolia and C. japonica. Non- Frankia actinobacterial isolates were also obtained from surface disinfected root nodules of C. myrtifolia and have been assigned to Streptomyces (n=5), Micromonospora (n=5), Verrucocispora (n=2), Micrococcus (n=1), Plantactinospora (n=1), and *Nocardia* (n=12). Results obtained in this study are confirmative to previously reported data showing low genetic variability, saprophytic potential of *Frankia* cluster 2 and lack of cospeciation with their respective host plant species.

# Heavy metals effect on two strains bacteria's growth isolated from contaminated ghouts of Oued Souf region

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# Abstract

Some south Algeria areas and particularly the region of Oued Souf are confronted with the phenomenon of the water rise which appears by an environmental pollution. The water rise made that the level of the surface of subsoil waters exceeds sometimes the level of the ground in the ghouts, characteristic of Oued Souf and which are depressions realized men, to make it possible the palm roots close to the ground water. The water rise phenomenon has as consequences deterioration of the ghouts and pollution of the deep ground water.

The objective of our work is to try to know more about mineral and microbiological pollution in ghout water, and also, to test the capacity of resistance to heavy metals (Cd, Pb and Cu) of certain bacterial strains isolated from the polluted biotopes. The results of the physicochemical analyses showed an apparent pollution in the sites. We could isolate around fifty bacterial strains belonging to different kinds, 5 fungal strains and 12 cyanobacterial and algae strains. Two bacterial strains, *Aeromonas* sp. and *Staphylococcus xylosus* were selected to be tested for their resistance to heavy metals (Cd, Pb and Cu). The study of their kinetic growth in medium added with metals, showed that *Aeromonas* sp. has resists to until 1000 mg/L of all the metals, Cu, Pb, Cd. Concerning *Staphylococcus xylosus* its growth clearly slowed down at 1000 mg/L of Cd concentration.

Pb and Cu the growth is significantly important with low concentrations of Pb and Cu, but was slowed down considerably with the high concentrations (1000 mg/L).

Therefore these strains could be used in the control of metal pollution, but other studies must be realized to understand their mechanisms of resistance.

Keywords: ghout, pollution, heavy metals, Aeromonas sp., Staphylococcus xylosus.

# Diversity of culturable halophilic archaea isolated from Oued Melghir Lake (Algeria)

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# Abstract

Extreme ecosystems present drastic conditions that are deemed to be difficult for all forms of life. Such ecosystems are many in the Algerian Sahara. Their operation allows exploration of the frontiers of life while giving another dimension to the biodiversity of the microbial world. The Oued Melghir Lake, the largest lake in Algeria, is an extreme environment localized in north-west Algeria. The physicochemical and biological properties of The Oued Melghir Salt Lake were examined. It is saline and alkaline, where Na<sup>+</sup>, Cl<sup>-</sup> and SO3 <sup>-</sup> are the predominant ions, with a pH ranging from 8.4 to 9.2 and electrolytic conductivities from 14.03 to 127.6 Ms/cm. We have studied the diversity of culturable halophilic Archaea at Oued Melghir soil. We isolated a total of 40 strains, the isolates were aerobic, non-motile, Gram-negative organisms and exhibited little capacity for fermentation. All of the isolates were halophilic, with most requiring at least 15% salinity for growth, and all grew at 30% salinity. The isolates were mainly mesothermic and could grow at alkaline pH (8.5), which 4 different genera of halophilic archaea were identified, with Halopiger being the most abundant. The results of this study are discussed in terms of the ecological significance of these microorganisms in the breakdown of organic matter in Lake Oued Melghir and their potential for industry applications.

Keywords: biodiversity, archaea, cultivation, halophile, Halopiger.

# The maldi-tof ms technology serving both microbial ecology and materials chemistry

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# Abstract

The <u>Matrix-assisted</u> <u>Laser</u> <u>Desorption/Ionisation</u> <u>Time-of-Flight</u> <u>Mass</u> <u>Spectrometry</u> (MALDI-TOF MS) is a technology allowing the ionization/desorption of peptides and proteins from whole bacteria vegetative cells or spores, fungi and viruses. This method is used to identify many unknown microorganisms accurately. Unlike conventional identification techniques, such as biochemical/ phenotypic tests or molecular characterization by gene sequence analysis, the MALDI-TOF MS is an alternative method that offers fast and inexpensive analysis and provides reproducible and reliable identification of the unknown studied microorganisms at the genus, species and even at the sub-species level. Furthermore, microbial detection, screening and identification are systematic processes to study the microbial phylogeny and biodiversity of a given ecosystem. Indeed, it is necessary to identify each element of the microbial population in order to understand how microorganisms can interact with their environment. In this work, we are interested in identifying some native strains that have adapted to their environment and presenting a biotechnological interest. However, our challenge is also to combine microbiology to materials chemistry field. This can be done by elaborating efficient, stable and recyclable bio-inspired or biomimetic materials, without altering the activity of the microorganism of interest. In fine, such tools will find applications in many areas such as those related to pollutants biodegradation or to the bioremediation of polluted environments (soil / water). In this strategy, isolation of a strain from the environment in order to clean up a given polluted environment seems to be interesting, so the full circle might be complete.

**Keywords**: Microorganisms identification, biotechnology, microbial ecosystems, mass spectrometry, biomaterials, environment, biodiversity.

# Diversity of culture-dependent halophilic archaea in melghir Sabkha

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# Abstract

Halophilic Archaea have been isolated from a variety of hypersaline environments such as saline lakes, saline soils, and ancient halite. They are classified into the domain *Archaea* and constitute a large proportion of this habitat where salinities range near saturation. Their adaptations to grow at high salinity concentrations make them interesting for fundamental research, development of biotechnological process and the use of halophilic in bioremediation and biodegradation of various materials from industrial effluent to soil contaminations are explored. The aim of this work was to describe the archaeal community retrieved from Melghir and bettioua sabkha using culture-dependent molecular approaches. The production of extracellular hydrolase was qualitatively studied on these isolates. Phylogenetic analysis of 16S rRNA gene sequences revealed that the archaeal isolates displayed close relationships to several species. They were placed within the family *Halobacteriaceae*, and belonged to the genera: *Haloarcula, Halogeometricum, Halomicrobium* and *Halorubrum*. The isolates were characterized by their obligate Halophilic lifestyle and their aerobic heterotrophic metabolism.

# Biodiversity of rock-dwelling actinobacteria isolated from different monuments and stones in Algeria

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# Abstract

Thirty one isolates were obtained on different media including GYM and R2A from different monuments and various stones in Algeria (Ain Saleh, Askrem, Afilel, Timgad, Malbou, Oued Labiadh, Route de Ain guezzem, Ain chebbi, mountain of Hogar and Zana el Beidha). The 16S RNA genes sequencing assigned the isolates to four different genera; *Geodermatophilus*, *Modestobacter* and *Blastococcus* (Geodermatophilaceae) and *Friedmanniella* (Nocardioidaceae). Phenotyping and chemotaxonomic analysis are currently undertaken for several of the isolates to be proposed as type strains for the description of several novel species.

# Screening of prokaryotic species involved in soft rot, isolated from tubers and fruits, collected from different northeastern Algerian arid areas: preliminary study

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# Abstract

Vegetables, by their wealth in protein, vitamins and minerals, provide a balanced diet. They are divided into leafy vegetables, seeds vegetables, fruiting vegetables and root vegetables (tubers). Among these: potato (Solanum tuberosum), carrot (Daucus carota), which are the most susceptible to soft rot. Various bacterial species are responsible for diseases, affecting tubers, causing losses in fields or during vegetables storage. The aim of study was identification and screening of bacterial species responsible for soft rot of tubers (vegetables), warning of 05 plant species, Potatoes, Carrots, Turnips, Beets, and Zucchini, collected from local markets (Bordj Bou Arreridj province) north-eastern of Algeria. Isolation, purification on 05 selectif or not selectif culture media (LPGA, KingaA, King B, Agar Citrimide, and nutrient agar), Bacteriological, biochemical characterization by conventional galleries, and API 20<sub>NE</sub> API 10<sub>S</sub> ApiStrip ranges (BioMerieu) and physiological by cultures on extreme environments at various pH, temperature, 5T°C-concentrations (NaCl) medium, cultivation on various medium. Allows the selection of eight potentially saccharolytic pectinolytic (in vivo) bacterial species. After Api web identification, (with logiciel) these bacterial species were: Erwinia spp, Erwinia carotovora, Pseudomonas sp (carrots), Pseudomons fluorescens, Enterobacter cloaceae, Pseudomonas sp (potatoes), Pasteurella sp (Turnips), Pseudomonas fluorescens from beet. Antibiogramme showed that: 78% of strains were resistant, 15% sensitive and 08% were intermediate.

Keywords: Tubers, Soft rot, Bacterial Species, Screening, Antibiogram.

# Diversity of arbuscular mycorrhizal fungi associated to the rhizosphere argan trees from north-western of Algeria

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# Abstract

The study aims to evaluate the diversity of arbuscular mycorrhizal fungi associated with the rhizosphere of argan trees (*Argania spinosa* L. Skeels) growing in the region of Chlef and that of Mostaganem. The identification of these fungi has been performed by microscopic analysis of stained roots by trypan blue. The isolation of spores was done by wet sieving. Observation of root system of the argan trees under the microscope revealed the absence absorbent pans; this character shows the dependence of the argan to mycorrhizal fungi for the absorption of minerals. The observation of root fragments taken from the two regions has helped to highlight the presence of arbuscular mycorrhiza. The study also reveal the identification of four mycorrhizal species, three species belong to the genus *Glomus aggregatum*, *Glomus constrictum* and *Glomus sp*) and one specie belongs to the genus *Gigaspora* (*Gigaspora maragrita*). The results of isolation and identification of spores are similar in both regions in terms of species and different types of side number of spores; Whereas the ecological factors such as climate, rainfall, temperature, and biotic factors: soil type, the phosphorus content, the floristic surrounding the tree trunk.

# Bacterial counts and characterization of caecal flora of broiler chicks

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# Abstract

The aim of this study was to count, isolate and identify bacteria colonizing the caecum of broiler chicks. At 21 d of age, chicks were slaughtered and their intestinal tracts were removed. Samples of fresh digesta (1g) from caeca were collected. Microbial populations were determined by serial dilution  $(10^{-1} \text{ to } 10^{-7})$  of samples in saline solution. Plate count agar, Mac Conkey agar and Mannitol Salt agar were used for the isolation and enumeration of total bacteria count, Enterobacteriaceae and Staphylococcaceae, respectively. For the enumeration of lactic acid bacteria Man, Rogosa and Sharpe agar was used. The cell morphology was determined microscopically after Gram stain preparation. Catalase and oxidase tests were also determined. Pure bacterial strains were identified using API system (API 20E and API Staph). Results showed that strains of lactic acid bacteria are the most abundant (8.37±0.16 log10 UFC/g) than enterobacteriaceae genera (6.33±0.06 log10 UFC/g) and staphylococcaceae (4.42±0.09). Biochemical identification has revealed that *Escherichia coli* represented nearly 80% of all enterobacteriaceae isolated from caecum. Moreover *Staphyloccoccus aureus* represented 60% of Staphylococcaceae isolates. Further studies at the molecular level are needed to clearly identify caecal microbiota of broiler chicks.

Keywords: Bacterial counts, caecal microflora, broiler chicks.

# Characterization of Extremely Halophilic *Archaea* Isolated from Saline Environment (Sebkha Sidi Ameur) in Algerian Sahara

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# Abstract

The increasing interest, in recent years, in microorganisms from hypersaline environments has resulted in the discovery of several new species and genera. Members of the Halobacteria class are the dominant microorganisms in hypersaline environments worldwide. The haloarchaeal diversity of Sebkha Sidi Ameur (Bou Saâda) located in north-central Algeria was investigated using convential microbiological and molecular biology methods. Ten halophilic archaeal isolates selected were taxonomically classified on the basis of their morphological, physiological, biochemical properties and 16S rDNA sequences. The 16S rRNA gene sequences of the 10 strains were amplified by using PCR determined by the DNA sequencer and analyzed through the BLAST program subsequently. The ribosomal data showed that the isolates belonged to 4 genera within the Natrialbacea and Haloferacaceae families with the dominance of Natrialba (6 Strains), Narinema (2 stains) Halopiger and Haloferax (1 strain each) All strains showed 99% sequence identity with validly described species. Phylogenetic tree reconstructions based on the sequences obtained and the sequences available in databases was constructed. All the strains grew best with around 25% w/v salts, required high concentrations of NaCl; they revealed high degree of similarity with the closest described species in terms of colony, cell morphologies and physiological characteristics

Keywords characterization, Sidi Ameur Sebkha, halophilic Archaea, Natrialba

# Diversity of rhizobacteria isolated from halophyte in three different Sebkha soils

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# Abstract

Microbial diversity studies are important in understanding the microbial ecology in soil and other ecosystems. The diversity of bacterial communities on the rhizospheric soil of the halophytes in the saline arid areas remains largely unknown. The aim of this work was to define and monitor the structure of bacterial populations in the rhizosphere of a halophyte plant, Salsola tetranda, found in three different sebkha soils (Oran, Baniou and Melghir) in Algeria. To have access to an independent cultivation molecular analysis, total community DNA was extracted from the bacterial pellet recovered from root samples. 16S rDNA fragments amplified by PCR from rhizosphere bacterium DNA were analyzed by Terminal-Restriction Fragment Length Polymorphism (T-RFLP). Differences and similarities between the plant rhizospheric bacterial communities were very apparent. The T-RFLP data revealed enrichment of operational taxonomic units (OTUs) closely related to Acinitobacteriales, Rhizobiales, Burkholderiales, Pseudomondales and Sphingomondales. Salt soil-specific TRFs (Alcaliginales and Halomonales) were also detected. These findings show that T-RFLP of 16S rRNA genes can be used to compare between the different rhizospheric microbial communities and gain information about the numerically dominant populations that are present.

Keywords: rhizosphere, microbial diversity, halophyte, saline soil.

# *Archaea*, an Overview from the NCBI Nucleotide Database Perspective, a Meta-Analysis on: Biogeography, Biodiversity, and Screening for Interactions and Activities

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# Abstract

Archaea have been fascinating microorganisms to study for their roles and for the varieties of habitats they occupy. In this meta-analysis study, the metadata of NCBI Nucleotide Database is the first time investigated for these organisms to determine their: i) worldwide biogeography and biodiversity over 256 geographical areas, ii) ecological involvement and interest for the scientific community iii) occurrence under different physical-chemical conditions and in natural and artificial habitats. This was achieved by cross-linking the records to 80 sets of descriptor keywords, which are defined to cover a range of: habitats (soil, water...), organisms (plants, bacteria...), conditions (thermophile, halophile...), materials (Iron, Hydrogen...), enzymes (lipase, topoisomerase...), active biomolecules (antibiotics, antitumoral...) and interactions (oxidizing, reducing...). Besides, the ratios Environmental /Archaea and Crenarchaeota/ Euryarchaeota and alpha diversity indices were calculated. Finally, the study traces the evolution of registration of archaea and the number of publications related. Data-mining was performed through web service using *E-utilities* and Python. 6541 unique sequences were selected from all the 161585 ones retrieved from this database. These were present only over 97 geographical areas and correlated with 70 sets of descriptor keywords of the initial lists. Euryarchaeota was the most abundant and distributed phylum. China was the most important contributors in terms of submitted sequences while using diversity indices it arrived behind other countries. 82.60% of all unique records of this study have been submitted since 2008, and 2013 was the most important submitting year. 839 publications with PubMed ID (PMID) were linked to at least one unique record.

Keywords: NCBI Nucleotide, Metadata, Data-mining, Archaea Biogeography, Archaea Biodiversity, Archaea Activities.

# Biodiversity of Actinomycetales bacteria isolated from Chott Tinsilt

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# Abstract

Currently, the extreme ecosystems represent a coveted research field aiming of interesting microbial species, among which actinomycetes, adapted to the extreme conditions which reign during the biotechnological processes of synthesis. With this perspective, an approach was adopted in order to isolate and study the biodiversity of extremophiles actinomycetales from an extreme ecosystem"ChottTinsilt" (Wet Zone of east Algeria, classified as Ramsar site in 2004). For this, three soil samples were taken. A physicochemical study (pH measurement, determination of the electrical conductivity and the salinity, moisture content, rate of the organic matter and particle size) of these samples was performed and revealed the following results: the soil samples are alkali (8.57), extremely salty (79.11meq/100g), sandy clayey (33.73 % -31.37%) with moderate organic matter (8.62%) and low moisture content (3.58%). An experimental protocol based on the use of ISP5 medium supplemented with 2, 5, 10 and 15% NaCl respectively was adopted or the isolation of slight, moderate and extreme halophilic actinomycetes, from the three samples. This protocol allowed us to isolate 42 strains of actinomycetes. The Molecular identification by amplification and sequencing of the gene encoding the ribosomal 16S RNA has allowed the classification of the isolated strains in four different genera: Streptomyces (dominant genus) with 59.52%, Nocarsiopsis (35.71%), Brevibacterium (2.38%) and Actinopolyspora (2.38%). All strains are halotolerant except one that is halophile. 83.33% of strains tolerate 10% of NaCl and 16.66% tolerate 15% of NaCl. Consequently, these bacteria present a sure biotechnological interest.

Keywords: Biodiversity, Actinomycetes, Halophillic, Halotolerants.

# The morphological and physiological diversity of Actinobacteria in Algerian hot springs

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# Abstract

In Algeria, there are extreme ecosystems such as sebkhas, desert soils and above all the hot springs exploited for their benefits, especially treatment, but have been little studied from their perspective's biodiversity. The distribution of these hot springs is very irregular, their number increases as we approach the East. In this work, we have investigated the Actinobacterial diversity of these springs. Actinobacteria were collected from samples of water and mud in five hot springs. Only four strains of Actinobacteria were isolated from the mud Hammam Charef (Wilaya of Djelfa). These strains were subjected to phenotypics, physiological and biochemical studies. The cultures of strains isolated in five agar media used (ISP1, ISP2, ISP4, ISP5 and ISP7) are characteristic of bacteria which belong to the class of Actinobacteria, and the order Actinomycetales. The bacterial strains were revealed filamentous, after observation of stained smears, Gram-positive coloring, therefore they are of the order Actinomycetales. According to the physiological and biochemical studies, the results obtained show that the strains have a more or less important metabolic biodiversity with the possibility to produce or degrade more substrates through the development of different enzymatic activities. The results open perspectives and it would be interesting to continue this work with chemotaxonomic study and molecular identification of these strains by sequencing of the 16S RNA to determine their exact phylogenies.

Keywords: Actinobacteria; hot springs; phenotypic and physiological study; biodiversity.

# The control assay of vascular wilt of chickpea by the combinaison of two methods

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# Abstract

In Algeria, the culture of chickpea occupies an important place. But, this culture is exposed into naturals infections. Among the most destructive pathogens Fusarium oxysporum f. sp. ciceri has a particular attention. This saprophyte pathogen infects the soils and causes the vascular wilt of chickpea. In this work, first, we are tested in vitro actinomycetes of soil antagonistic against 03 isolates of Fusarium oxysporum f. sp. ciceri. Then the actinomycete the most antagonistic has been later tested in vivo with combination of three (ILC-482, PPC-25, Col-27) chickpea varieties from susceptible to moderate of vascular wilt and two moderate varieties. We have isolated 61 actinomycetes from arid soils of south Algeria presented different morphological characters. The antagonistic activities tests of these isolates by diverse methods had allowed us to select 6 isolates, presented interesting antagonistic activities against 3 isolates of *Fusarium oxysporum* f. sp. ciceri. The results of this test have showed that the isolate ACT2 has the most important activity. The results obtained in vivo have showed that the degrees of disease suppression by the actinomycete ACT2 vary. These degrees vary from 00 to 21.2% amongst ILC- 482 variety, from 00 to 27.78% amongst Col-27variety and from 12.58% to 24.55% amongst PPC-25 variety. The maximum degree of disease suppression has been observed amongst Col-27variety. The isolate ACT2 has presented high degrees of the disease suppression with the moderate varieties Col-27 (13.68%) and PPC-25 (18.78%) than the susceptible variety ILC-482 (8.76%).

Keywords: Fusarium oxysporum f. sp. ciceri, Cicer arietinum L., biocontrol, vascular wilt, actinomycetes, antagonism.

# Isolation of actinomycetes strains produce bioactive substances from Soummam Watershed sediments and their biodegradation ability of crude oil

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# Abstract

The impact of the increase in treatment failures of the bacterial resistance to classical antibiotics is increasingly important. It is therefore necessary to discover new bioactive molecules. Bioremediation is a recent technique, which is the voluntary use of spontaneous biological agents to remove or inactivate pollutants from soil, subsoil, groundwater and air. The aim of this work is to isolate antibiotic producing actinomycetes strains from the sediments of Soummam watershed and to study biodegradation ability of crude oil by these bacterial isolates. The physicochemical properties of Soummam Watershed sediments indicated that it was clayey, neutral, slightly salty sediment and that having high level of organic matter. 16 strains of actinomycetes were isolated from sediment Watershed Soummam. Antimicrobial activity was tested against three Gram-positive bacteria and three Gram-negative bacteria, one yeast and two filamentous fungi, using the agar cylinder method. Among these isolates, 06 strains showed an activity against at least one of the germ-tested. The isolate SSB<sub>2</sub> showed antibacterial and antifungal activities. To select of actinomycetes strains degrading crude oil, enrichment cultures on basal mineral salt medium supplemented with crude oil as unique carbon and energy source were performed. The confirmation of the biodegradation ability of crude oil was tested on basal mineral salt medium agar by hole-plate diffusion method, 10 actinomycetes strains were able to degrading the crude oil on the basal mineral salt medium broth and agar.

**Keywords:** Antimicrobial activity, agar cylinder method, biodegradation, crude oil, holeplate diffusion method.

# Identification and technological characterization of lactic acid bacteria isolated from wheat flour

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# Abstract

Cereals that are well known to be a historic base of the Mediterranean diet, still occupy today a prominent place in agriculture and agri-food production in Tunisia, as well as the household food consumption. Bread is one of the staple foods consumed by humankind. In order to improve its quality, the industrialists use sourdough that contains complex cultures of symbiotic yeasts and lactic acid bacteria (LAB). The study has focused on 20 baking flour samples collected from 13 mills spread throughout the entire Tunisian country. The sanitary conditions of the analyzed samples were assessed through the enumeration of total microflora, lactic acid bacteria, yeasts and molds. Moreover, we focused on isolation and characterization of LAB at the molecular and the technological level. In total, 228 strains have been isolated by using Man Rogosa Sharpe (MRS) medium, supplemented with the bromocresol green. Only 47 Gram- positive and negative-catalase isolates were retained. The selected strains were subjected to DNA extraction and PCR amplification of the intergenic spaces (ITS). Based on the analysis of ITS profiles, 13 different haplotypes were distinguished and compared. The selected isolates were assessed for acidifying activity antibacterial activity against Listeria monocytogenes, Staphylococcus aureus, Escherichia coli and Enterococcus faecalis. The obtained results revealed that most of the strains showed interesting and remarkable acidifying activities up to pH 3.8. Moreover, they were able to inhibit the growth of S. aureus, and with a lesser extend E. faecalis and E. coli.

# Comparative study of monogeneans parasites of two populations of cyprinids (*Barbus callensis* and *Cyprinus carpio*) populating the Dam Foum El-Khanga (Souk-Ahras)

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# Abstract

The comparative study of the monogeneans parasites community of 224 individual fish of two freshwater species in the family Cyprinidae: *Barbus callensis* and *Cyprinus carpio* populating the dam Foum El-Khanga in Souk Ahras during for four months (January-April 2015) resulted in isolation of 1444 monogeneans gill. Observation of morphological and anatomical characteristics of the isolated parasites led to identified ten species of Monogenea sub Monopisthocotylea class belonging to the genera: *Dactylogyrus Pseudacolpenteron* and seven species found at *B. callensis* (*D. extensus, arcuatus, ksibii prostae, heteromorphus, borjensis, P. Pavlovskii*) and six in *C. carpio* (D. *extensus, vastator, arcuatus, anchoratus, formosus, P. Pavlovskii*). The results of the distribution of species monogeneans in both host species, showed the presence of certain parasites permanently during the four months of study: *D. heteromorphus, D. Prostea, P. Pavlovskii* (in *B. callensis*) and *D. extensus, vastator, P. Pavlovskii, D. anchoratus* (in *C. carpio*), the rest of parasitic species: *D. extensus, and D. ksibii borjensus* was present only during the spring period in *B. callensis*, while *D. arcuatus* species occurred only in winter in both host species.

Key words : Barbus callensis, Cyprinus carpio, Dam Foum El Khanga, Monogenea, season.

# SUMMARY

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